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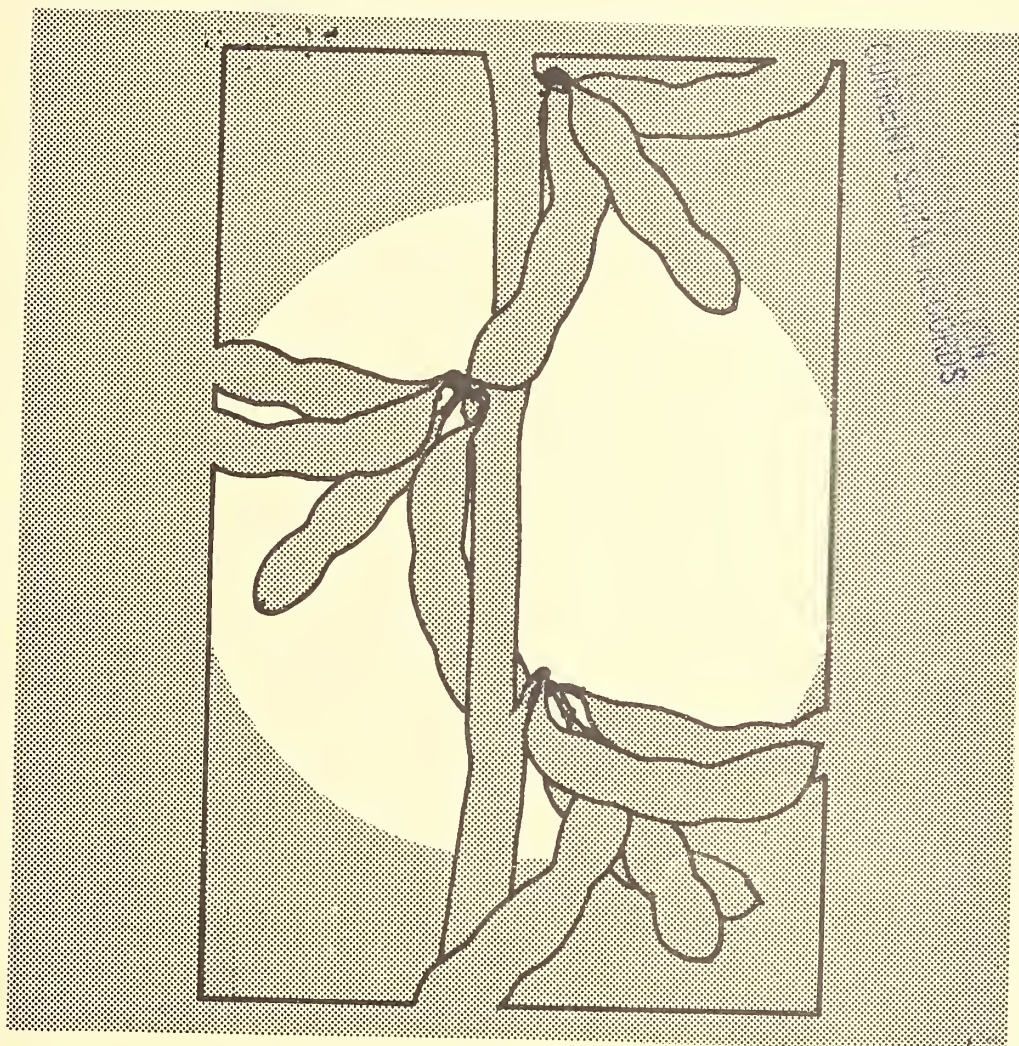
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RESERVE

# Soybean Genetics Newsletter



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Volume 9

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Agricultural Research Service - USDA  
Department of Agronomy  
and Department of Genetics  
Iowa State University  
Ames, Iowa 50011





## TABLE OF CONTENTS

	page
I. FOREWORD . . . . .	1
II. ANNOUNCEMENT . . . . .	2
III. QUARANTINE RESTRICTIONS . . . . .	3
IV. RSCRE MEETING REPORT . . . . .	4
V. REPORT OF THE SOYBEAN GENETICS COMMITTEE . . . . .	9
VI. USDA SOYBEAN GERMPLASM REPORT . . . . .	15
VII. RESEARCH NOTES	
<u>Brazil:</u>	
Reaction of soybean cultivars to Temik (aldicarb) and inheritance of the reaction. R. A. S. Kiihl and L. A. Almeida . . . . .	17
Calico mosaic of soybeans: Sources of resistance and inheritance of reaction. A. M. R. Almeida, R. A. S. Kiihl and L. A. Almeida . . . . .	18
<u>Canada:</u>	
Soybean linkage and crossover tests. R. I. Buzzell and R. D. Walker . . . . .	23
Inheritance of presence/absence of flavonoid compounds in soybean seedcoats. R. I. Buzzell and B. R. Buttery . . . . .	24
Genetics of black pigmentation of soybean seedcoats/hila. R. I. Buzzell and B. R. Buttery . . . . .	26
Soybean emergence in clay soil and tolerance to phytophthora rot. R. I. Buzzell and T. R. Anderson . . . . .	29
Greenhouse determination of soybean tolerance to phytophthora rot. R. I. Buzzell, C. G. Mortimore and J.H. Haas . . . . .	30
<u>India:</u>	
'N-23-A' A new promising variety of soybean for Chotanagpur. H. B. P. Trivedi and R. Prakash . . . . .	33
Effect of growth regulators Cycocel (CCC) Regim-8 (TIBA) and Ethrel (CEPA) on soybean crop. M. S. S. Rao, P. C. Agrawal and R. Prakash . . . . .	35
New breeding lines of soybean developed at Pantnagar. H. H. Ram, V. D. Verma, K. Singh and Pushpendra . . . . .	39
Extent of selfing during crossing in soybean. H. H. Ram, V. D. Verma, Pushpendra and K. Singh . . . . .	42
Screening for photoperiod insensitivity under natural conditions in soybean. H. H. Ram, K. Singh, Pushpendra and V. D. Verma . . . . .	43
Genotype X Environment interaction in soybean: I. Individual regression analysis. V. P. Gupta, I. K. Garg and N. D. Rana . . . . .	45
Genotype X Environment interaction in soybean: II. Joint regression analysis. V. P. Gupta, I. K. Garg and N. D. Rana . . . . .	47
Association among productivity, responsiveness and stability for different groups of traits in soybean. V. P. Gupta, I. K. Garg and N. D. Rana . . . . .	51

Genetic control of productivity, responsiveness and stability for different groups of traits in soybean. V. P. Gupta, I. K. Garg and N. D. Rana . . . . .	53
Factor analysis in F <sub>2</sub> generation of soybean crosses. S. K. Sharma, B. M. Ashawa and N. D. Rana . . . . .	58
Effect of environment and cropping system on the coefficients of variability for seed yield, quality, structural, phenological and physiological traits in soybeans. V. P. Gupta, I. K. Garg and N. D. Rana . . . . .	62
Consistency of heritability estimates over environments and cropping systems for different groups of traits in soybean. V. P. Gupta, I. K. Garg and N. D. Rana . . . . .	66
Association of leaf and root characteristics among themselves and with seed yield, structural, physiological and phenological traits in soybean. V. P. Gupta, I. K. Garg and N. D. Rana . . . . .	68
Variation and heritability for leaf and root characteristics in soybean, across locations. V. P. Gupta, I. K. Garg, N. D. Rana and J. M. Singh . . . . .	71
Genetic, altitude and climatic effects on seed yield and germinability traits in soybean. V. P. Gupta, N. D. Rana, R. K. Sharma and G. Chand . . . . .	75
Correlation among seed yield, seed quality and nutritional traits in soybean. N. D. Rana, R. K. Kalia and V. P. Gupta . . . . .	81
<u>Nigeria:</u>	
Constraints in using seed leachate characteristics to estimate seed vigor for varietal seed keeping quality comparisons in soybeans. G. Gumisiriza and E. A. Kueneman . . . . .	87
<u>Taiwan:</u>	
Relationship between photoperiod, temperature, solar radiation and grain yield in soybean. S. Shanmugasundaram and C. R. Yen . . . . .	93
Screening for immature green soybeans as a vegetable. S. Shanmugasundaram and C. R. Yen . . . . .	95
Yield evaluation of immature, green soybeans. S. Shanmugasundaram, T. S. Toungh and R. B. Almodiente . . . . .	97
Forcing soybeans to mature by spraying paraquat. S. Shanmugasundaram and T. S. Toungh . . . . .	99
<u>Thailand:</u>	
A second report on induced mutations for soybean rust resistance. S. Smutkupt, A. Wongpiyasatid and S. Lamseejan . . . . .	103
<u>United States:</u>	
The effects of temperature on longevity and vitality of soybean seeds. Z. E. Bailey . . . . .	109

Phytoestrogens in wild perennial relatives of the soybean. D. A. Vaughn and T. Hymowitz . . . . .	112
Root fluorescence in the Genus <i>Glycine</i> Subgenus <i>Glycine</i> . C. Parot . . . . .	115
Genetic analysis of a chlorophyll deficient, tan-saddle mutant. R. C. Shoemaker and R. G. Palmer . . . . .	117
A duplicate-deficient line in soybeans. K. Sadanaga and X. Delannay	121
A dwarf mutation in 'Hodgson' soybean. K. Sadanaga and R. Grindeland . . . . .	123
Chlorophyll-deficient plants in a soybean cross. K. Sadanaga . . .	126
Identifying translocations in soybeans. K. Sadanaga and K. Newhouse	129
Genetic linkage analysis. T. E. Devine and B. H. Breithaupt . . . .	131
The genetic basis of physiologic races of phytophthora. V. D. Leudders . . . . .	135
Preliminary electrophoretic observations from several soybean enzymes. M. B. Gorman, Y. T. Kiang, Y. C. Chiang and R. G. Palmer . .	140
Electrophoretic classification of the early maturity groups of named soybean cultivars. M. B. Gorman, Y. T. Kiang, Y. C. Chiang and R. G. Palmer . . . . .	143
A somatic approach to soybean genetics. J. Roth and K. G. Lark . .	157

#### U.S.S.R.

Protein content in grain and lysine content in protein of soybean mutants, induced by chemical mutagens and gamma rays. V. I. Sichkar, A. P. Levitsky and V. F. Marjushkin . . . . .	161
The lectin content in different varieties of soybean in connection with improving nutritive qualities. E. L. Golynskaya, M. V. Kovalchuk, V. I. Sichkar, V. F. Marjushkin and A. P. Levitskij . . .	163

#### Vietnam

Decapitation technique to increase grain yield in soybean. C. H. Tin . . . . .	167
Nontillage is a technique in growing soybean. T. T. Tuan and C. H. Tin . . . . .	168
Exotic soybean observational yield performance trial in Kien Gian province -- Mekong Delta -- Vietnam -- dry season 1981-1982. N. V. Thieu, C. H. Tin, T. V. Hiep, V. Lau and V. C. Khanh . . . . .	169

#### Zambia

Notes on soybean nodulation with the indigenous <i>Rhizobium</i> in Zambian soils. F. Javaheri and R. Nyemba . . . . .	173
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VIII. INDEX OF AUTHORS . . . . .	175
IX. RECENT SOYBEAN GENETICS AND BREEDING PUBLICATIONS . . . . .	176
X. MAILING LIST . . . . .	187



## I. FOREWORD

In the nine years since its inception, the Soybean Genetics Newsletter has grown in many ways. We've been pleased and proud of the response of soybean scientists all over the world. It is apparent from the volume, quality, and scope of the articles submitted, and from the number of "subscribers" that our newsletter is a valuable tool in communication among researchers.

There has come the time, however, when we must establish some guidelines, to keep this volume from overwhelming us. It is not an easy task; no one person can say which article, what particular topic, whose research, is of most value and interest to other researchers. We felt it necessary, therefore, to discuss the subject of editorial guidelines with the Soybean Genetics Committee. We quote a paragraph from the minutes of the 22 February, 1982, meeting of the Soybean Genetics Committee:

"Dr. Palmer, as editor of the Soybean Genetics Newsletter, had asked the committee to discuss the procedure he might follow where many manuscripts are received from one author or institution..." on essentially the same research, when the same set of data are reprocessed from year to year...." Such authors might wish to consolidate several papers into a few, submit only their highest priority papers, or in other ways be selective of their contributions."

For Volume 10, in 1983, it is our intention to implement the suggestions of the Soybean Genetics Committee. If we receive several articles from one author, on essentially the same research, we will write and suggest that the author condense or combine the articles. Instead of trying to publish long lists of data, we will suggest that the author place a notation in his paper that readers wishing complete data can get them by corresponding directly with that author.

This year's newsletter, Volume 9, has been published with the able and willing assistance of graduate students Randy Shoemaker, Peggy Hatfield, Long-Fang Chen and Jeff Griffin, and technicians Sally Pyle and Holly Heer. Without them there would be no newsletter.

Reid G. Palmer, editor

The data presented in the Soybean Genetics Newsletter are not to be used in publications without the consent of the respective authors.

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## II. ANNOUNCEMENT

New Officers of the Executive Committee of the  
Commercial Plant Breeders Committee of  
National Council of Commercial Plant Breeders

## Chairman

Wayne Ellingson  
North American Plant Breeders  
RR #2, Hwy 30 East  
Ames, IA 50010

## Secretary/Treasurer

John Hicks  
Pioneer Hi-Bred Int'l.  
P. O. Box 4428  
Greenville, MS 38701

## Vice Chairman

John Schillinger  
Asgrow Seed Company  
634 E. Lincolnway  
Ames, IA 50010



PLANT INTRODUCTION OFFICE  
 Germplasm Resources Laboratory  
 USDA Agricultural Research Center  
 Beltsville, MD 20705

1) Quarantine restrictions protect against introduction of rust with soybean importations

Quarantine restrictions have been imposed on the importation of soybean seeds into the United States from certain countries because of the risk of introducing soybean rust (*Phakopsora pachyrhizi*). Seeds of *Dolichos*, *Pachyrhizus*, *Phaseolus*, *Pueraria*, and *Vigna* are also included in the quarantine regulation. Regulated countries are:

Eastern Hemisphere - Africa, Australia, Burma, Cambodia, India, Indonesia, Japan, Korea, Laos, Malaysia, Nepal, New Caledonia, Papua New Guinea, Peoples Republic of China, Philippines, Republic of China (Taiwan), Sri Lanka, Thailand, USSR, Vietnam;

Western Hemisphere - Brazil, Costa Rica, Venezuela, West Indies.

Plant Quarantine regulation CFR 319.37 requires the seed to be treated with Patterson's Multipurpose Fungicide (mixture of 21% zineb and 22% captan) dust at a rate of 1.05 oz or slurry at a rate of 0.74 oz per bushel of seed. Remember, the quarantine regulation may change especially with distribution shift of the causal organism.

For small experimental quantities of seed (total weight of 3 lbs or less per shipment), proper treatment can be handled at USDA's Plant Germplasm Quarantine Center (PGQC). Since the Plant Introduction Office (PIO) routinely monitors (in consultation with crop germplasm curators) plant materials that come through PGQC, importers of soybean seed might be asked to share the seed with the Soybean Germplasm Collection. If you need assistance or have questions about the introduction and exchange of experimental quantities of soybean seed, write to the authors.

Persons who wish to import soybean seed for propagation or other purposes should contact the Animal and Plant Health Inspection Service (APHIS) to work out permit arrangements. Contact Permit Unit, APHIS, U.S. Department of Agriculture, Federal Building, Room 638, Hyattsville, MD 20782.

For a general but informative article about soybean rust, see Intsoy Research Highlights, Intsoy Newsletter No. 27, November, 1981. Write to: INTSOY, Univ. of Illinois at Urbana-Champaign, 113 Mumford Hall, 1301 W. Gregory Drive, Urbana, IL 61801.

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Sharon Kenworthy

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Research on Soybeans for Cooler Regions of Europe  
Meeting at Eischikon near Zürich, 3./4.8.1981 [1,2]

> Notes on the meeting:

1. Introduction

Prof. E. R. Keller chaired the meeting and welcomed the participants. He specially mentioned Dir. M. Arnoux, chairman of the FAO-Soybean Network, Prof. Dr. G. Röbbelen, chairman of the Oil and Protein Crops' Section of EUCARPIA and Prof. Dr. W. D. Beversdorf, Univ. of Guelph (Canada), at present time guest scientist in the Crop Science Dept. at Zürich. Both organizations suggested to organize this meeting and the aim should be to: create a working group of specialists which, based on already available experience, should 1) encourage better cooperation in research, and 2) continue research on the adaptation of the soybean plant to cooler regions in Europe.

2. List of participants

<u>Canada</u>	Dr. W. D. Beversdorf, Dept. of Crop Science, Univ. of Guelph, Guelph, Ontario, N1G 2W1
<u>England</u>	Mr. S. Houghton, ADAS, Ministry of Agriculture, Fisheries and Food, Great Westminster House, Horseferry Road, London SW1P 2AE
<u>France</u>	Mr. M. Arnoux, Ecole Nat. Sup. Agron., Dir. Stat. d'amélioration des Plantes, F-34060 Montpellier-Cedex Mr. A. Vidal, Station d'amélioration des Plantes INRA, F-34060 Montpellier-Cedex
<u>Federal Republic of Germany</u>	Miss I. Kübler, Justus Liebig Universität, Institut f. Pflanzenbau u. Pflanzenzüchtung, D-6300 Giessen (Prof. Dr. W. Schuster) Prof. Dr. G. Röbbelen, Institut f. Pflanzenbau u. Pflanzenzüchtung, V.-Siebold-Strasse 8, D-34 Göttingen Dr. J. F. Seitzer, Dir. Inst. f. Pflanzenzüchtung, KWS Kleinwanzlebener Saatzucht, D-3352 Einbeck Dr. A. Tékéte, Univ. Hohenheim, Institut f. Pflanzenbau u. Pflanzenzüchtung, Postfach 106, D-7 Stuttgart 70 (Prof. Dr. G. Kahnt) A. Adetoro, Univ. Hohenheim, Institut f. Pflanzenbau u. Pflanzenzüchtung, Postfach 106, D-7 Stuttgart 70
<u>Poland</u>	Prof. Dr. J. Szyrmer, Plant Breeding and Acclimatization Institute, IHAR, Radzikow near Warsaw, 05-870 Blonie
<u>Sweden</u>	Mr. R. Elovson, Holmbergs Utsaeden, Fiskeby, S-605 90 Norrkoeping

Switzerland Miss S. Uehlinger and Dr. W. Gehrig, Station Fédéral de Recherches Agronomiques, 1260 Changins-Nyon

Mr. W. Huber, Eidg. Forschungsanstalt für landw. Pflanzenbau, 8046 Zürich-Reckenholz

Prof. Dr. E. R. Keller, Mr. H. Brenner, Dr. J. Schmid and Dr. A. Soldati, Institut f. Pflanzenbau, ETH-Zentrum, 8092 Zürich

### 3. Papers presented

- R. Elovson: Soybean breeding in Sweden. I. The present position.
- A. Vidal: Soybean research and breeding program in INRA - Montpellier.
- J. F. Seitzer: Adaptation of soybeans to the growing conditions of Western Europe.
- J. Szyrmer, A. K. Szczepanska: Screening of soybean genotypes for cold tolerance during germination.
- W. D. Beversdorf and D. J. Hume: Cold tolerance research in soybeans.
- E. R. Keller, A. Soldati, J. Schmid, H. Brenner and W. D. Beversdorf: Adaptation of soybean to cooler regions of Europe.
- W. D. Beversdorf and D. J. Hume: Future effects for soybean improvement in Ontario, Canada.
- R. Elovson: Soybean breeding in Sweden. II. Plans for the future.
- A. Vidal, M. Arnoux: Proposals for a cooperation program "Adaptation of the soybeans to cooler regions."
- A. Soldati, E. R. Keller: Considerations on future research activities.
- H. D. Voldeng: Short season soybean program.

Summary: Discussion of the position of soybean research for cool regions of Europe (the full text of all papers was made available to all participants prior to the meeting).

Considerable time was devoted to establishing the current position of soybean research in the cooler regions of Europe. Objectives of the programs were consistent to the extent that all programs are attempting to increase yield and yield stability. Specific goals and approaches of the breeding programs varied considerably.

In France, Mr. Vidal reported that efforts are concentrated on developing group I and II soybean cultivars with improved drought tolerance for non-irrigated production areas and varieties with improved lodging resistance and reduced leaf area for irrigated fields. For the northern half of France, early varieties are required that have productivity characteristics of mid-west American varieties combined with cold tolerance characteristics of the Swedish varieties. In the Paris-basin the introduction of the soybean into rotations rich in cereals would be appreciated. A yield level of 2.5 t/ha (South) or 2 t/ha (North) is considered as competitive. In Montpellier, a cooperative effort with private companies and ENSAT was initiated in 1981 using a recurrent (3-year) selection scheme to develop adapted early soybean material.

In Poland soybean research has an old tradition. Soybeans are included in the National Plant Protein Research Program. Professor Szyrmer indicated that soybean breeding is concentrating on very early maturing (000-type) determinate material. Improved seedling emergence at low temperatures is a primary goal of the research and breeding efforts as well as improved quality characteristics related to human consumption of soy-protein products. In his breeding program he is working with the pedigree method as well as with mutagenesis. He emphasizes the necessity of physiological research work.

In Sweden, Mr. Elovson reported that breeding efforts are centered on improved yield and harvestability, and improved emergence at low temperatures. Sweden, like Poland, is concerned primarily with improvement of 'Fiskeby V' maturity material. Mr. Elovson indicated that improved lines may be available commercially in two years. He does not have a definite opinion in relation to the preference of determinate or semideterminate types.

Dr. Seitzer of the Federal Republic of Germany reported that yield and yield stability, cold tolerance during pod filling, resistance to *Pseudomonas glycinea* and improved seed quality are required in cool regions of Europe. Dr. Seitzer suggested that, while considerable genetic variability exists for yield improvement, variability for earliness and cold tolerance is somewhat limited.

Dr. Soldati of ETH, Switzerland, placed emphasis on the need to establish an ideotype for cooler regions. He indicated that agronomic practices, genotype and the environment all have a major influence on the development of the soybean plant and that the interaction of these basic sources of variation results in considerable variation of plant development from year to year and location to location. Dr. Soldati indicated that improved cold tolerance, particularly during flowering and pod filling, was necessary in early varieties for cooler regions of Europe.

Dr. Beversdorf of Ontario, Canada, suggested that temperature variation from year to year during critical developmental stages for soybean has led to yield and maturity instability in many cooler regions. He suggested the need to evaluate the natural environmental variability within each region of potential soybean production from historical weather data and the response of genotypes to this variation as the first step in defining requirements of adapted varieties and selection criteria that will ultimately lead to stable and adapted varieties for cooler regions of Europe.

The general approach to breeding improved varieties for cooler regions varied considerably among institutes, from mutation breeding and pedigree advance in Poland, to modified single-seed descent advance and recurrent selection schemes in France. The concept of cold tolerance also varied considerably among institutes from emergence at cool temperatures in Poland and Sweden to podding ability at low temperatures in West Germany and Switzerland. Evidence of the independence of specific cold-tolerance traits provided in the position papers resulted in general agreement that current varieties may have some cold-tolerant characteristics (e.g., ability to emerge at low temperatures) while lacking others (e.g., ability to set pods at low temperatures).

The relationship of photoperiod sensitivity and temperature on the duration of vegetative and reproductive stages was also discussed. There was general agreement that these relationships need further evaluation across the diverse ranges of photoperiods and climatic conditions of Europe. The North



American maturity system does not appear to adequately define maturity adaptation of varieties in cooler regions of Europe, particularly in regard to the very early groups (Group 0 and 00 of the North American system).

Discussion of desired plant types regarding termination of vegetative growth (degree of indeterminate habit) was discussed but conclusions varied. The desirability of a determinate habit with regard to uniform maturation was offset by the desirability of the indeterminate habit regarding recovery from stress (cool temperatures or leaf diseases) during critical early reproductive stages and harvestability. There was agreement that soybean has excellent ability for total biomass production in cool regions, but that instability in partitioning biomass between vegetative and reproductive growth was a problem. This partitioning is related to both temperature-photoperiod interactions and podding ability following cold stresses during early reproductive development, and therefore is subject to the highly variable environment of cooler regions of Europe. Genotypes with increased stability in this regard would facilitate development of agronomic practices that consistently provide maximum performance.

#### Priority areas of soybean - Research and cooperative efforts

Several participants provided suggested areas of research that might assist in the adaptation of soybean to cooler areas of Europe. General agreement on the need to evaluate the photoperiodic and temperature responses of current varieties led to the formation of a cooperative trial of a standard set of early cultivars across much of the cooler regions.

In order to allow integration of the results of these trials, minimum data collection criteria were established, as follows:

- a) Daily minimum and maximum temperatures will be provided from the date of planting until maturity.
- b) Daily or weekly precipitation will be recorded.
- c) Precise latitude and elevation of the trial locations will be provided, as well as soil type.
- d) General agronomic practices including seeding rate, row width and planting depth and date will be provided.
- e) Final emergence of plant density will be provided.
- f) The date of R<sub>1</sub>, R<sub>3</sub>, R<sub>5</sub>, R<sub>7</sub> and R<sub>8</sub> (according to the system proposed by Fehr and Caviness, 1977) will be recorded as precisely as possible (see discussion of the system below).
- g) The date of the first killing fall frost will be recorded if any variety in the trial has not achieved R<sub>8</sub> by that date.
- h) The date and type of any unusual weather or environmental event that might affect measurements of adaptation (hail, epidemic, nutrient deficiency, lodging due to wind storms, etc.) will be recorded.
- i) The trial will include a standard set of 8 to 10 early varieties subject to availability of quality seed. Seed will be distributed by Dr. Soldati, ETH.
- j) The cooperative trial will be conducted by participants for a minimum of two years beginning in 1982.

- k) Other data to be collected by cooperators include: Yield ( $\text{g/m}^2$ ), lodging (0 = no lodging to 5 = completely lodged), height (cm) and 1000-seed weight).

Several suggestions for other areas of cooperative research were proposed. Variation among institutes and availability of resources led to the conclusion that cooperation would be at the discretion of individual research groups. The priority areas identified included:

- a) Evaluation of the current extent of variability for specific cold tolerance traits, early flowering and early maturity and estimating the heritability of desirable characteristics.
- b) Assessment of the need for and potential benefits from a potential FAO-sponsored collection of early material from North Japan, North China and the Eastern USSR some time in the near future (in this regard, a subcommittee was established to evaluate the currently available sources of very early maturing entries in various germplasm collections; Dr. Seitzer, responsible with Prof. Beversdorf, Mr. Elovson, Prof. Szyrmer and Mr. Vidal).
- c) Exchange of breeding materials (i.e., from SSD programs and material that appears productive but out of the specific area of adaptation for which the originating program is concerned).
- d) Exchange of early maturing sources of resistance to *Pseudomonas glycinea*.
- e) Evaluation of the effects of different planting dates within locations for lengths of developmental stages for the standard set of cultivars used in the cooperative trial above.
- f) Evaluation of the responses of the standard set of cultivars (above) to different controlled photoperiods and different controlled temperatures with regard to duration of critical developmental stages ( $V_c$  to  $R_1$ ,  $R_1$  to  $R_3$ ,  $R_3$  to  $R_5$ , and  $R_5$  to  $R_7$ ), in order to establish ratings on photoperiod sensitivity which could be compared with estimates deducted from the cooperative trials.

#### Discussion on identification of growth stages:

Considerable variation in collection of data regarding the stage of development of soybean was apparent from institute to institute. In order that data from cooperative trials may be combined in developing models of soybean adaptation, the group decided to use the system proposed by Fehr and Caviness, 1977. A copy of the proposed system will be provided to participants of the cooperative trials (subject to permission of the authors) by Dr. A. Soldati.

#### Future activity

The participants elected Dr. A. Soldati as chairman of the working group. We will contact the different research institutes and ask them whether or not they wish to join the working group. He will serve as a coordinator for the above mentioned program and initiate it.

W. D. Beyersdorf  
 100 A. Soldati  
 E. R. Keller



245  
 N. REPORT OF THE SOYBEAN GENETICS COMMITTEE [1-2].

- A) The current members of this committee and the expiration dates of their terms are as follows:

100  
 R. L. Buzzell (1984)  
 Canada Dept. of Agriculture  
 Research Station  
 Harrow, Ontario  
 Canada NOR 1G0

H. H. Hadley (1984)  
 Turner Hall  
 Department of Agronomy  
 University of Illinois  
 Urbana, IL 61801

E. T. Gritton, Chm. (1983)  
 Department of Agronomy  
 University of Wisconsin  
 Madison, WI 53706

C. Newell (1983)  
 Turner Hall  
 Department of Agronomy  
 University of Illinois  
 Urbana, IL 61801

J. H. Orf (1985)  
 Department of Agronomy  
 University of Minnesota  
 St. Paul, MN 55108

R. G. Palmer, USDA, Ex Officio  
 (Editor of Soybean Genetics Newsletter)  
 Department of Genetics  
 Iowa State University  
 Ames, IA 50011

T. C. Kilen (1985)  
 Res. Geneticist  
 Soybean Production Research  
 P. O. Box 196  
 Stoneville, MS 38776

- B) Organization of the Committee:

- 1) The Committee will be composed of six elected members and the editor of the Soybean Genetics Newsletter.
- 2) The term of the elected members will be three years. After a member has been off for one year, he (she) can be reelected. The Committee will elect two new members each year; a simple majority is needed for election. The members will be elected prior to February 1 of each year, by a mail ballot conducted by the chairman.
- 3) At the annual meeting of the Committee (usually in February in conjunction with the Soybean Breeding and Genetics Workshop), the two new members and the two retiring members of the Committee are eligible to attend and vote.
- 4) The chairman will be elected at the annual Committee meeting and serve through the next annual meeting, and may be reelected.

C) The duties of this Committee include the following:

1) Maintain Genetic Collection.

The Genetic Collection is divided into four categories:

- a) Type Collection includes all published genes of soybeans, preferably in the original strains (excluding U.S. and Canadian name varieties, which are maintained in a separate collection) plus certain mutants or strains that appear to the Committee to have potential genetic interest.
- b) Isoline Collection includes adapted varieties Clark, Harosoy and Lee, into which have been backcrossed single genes or combinations of genes. Also included are certain genes or combinations with Chippewa, Wayne and Williams.
- c) Linkage Collection includes linkage combinations and the various genetic recombinations.
- d) Cytological Collection includes translocations, inversions, deficiencies, trisomics, tetraploids, etc.

Collections a, b, and c are maintained at Urbana, Illinois, with R. L. Bernard as curator. Collection d is maintained at Ames, Iowa, with R. G. Palmer as curator.

2) Manuscript review and genetic symbol approval.

The Soybean Genetics Committee requests that researchers submit all manuscripts concerning qualitative genetic interpretation and symbols to the Committee Chairman. This review by the Genetics Committee will serve to insure orderly identification and use of genetic nomenclature and to avoid conflict of symbols. This will also allow assignment of type collection designations (T-numbers) prior to publication, so that these T-numbers may be used in the journal article to identify parental lines.

3) Soybean Genetics Newsletter notes.

All notes for the Newsletter should be sent to the SGN editor, R. G. Palmer, who will ask the Soybean Genetics Committee to review those articles concerning qualitative genetic interpretation and symbols. Genetic symbols reported in the Newsletter will have the same status as those published in scientific journals.

D) The Committee will take the responsibility for publishing every five years, starting in 1983, in the SGN a list of all gene symbols, linkage groups, translocations, and trisomics in soybeans. Researchers who have references on the gene symbols and linkage groups are urged to send them to R. L. Bernard. Researchers who have references on translocations and trisomics are urged to send them to R. G. Palmer.

E) The function of the Committee was officially expanded to include genetics research in the entire *Glycine* genus rather than restricting its responsibilities to *Glycine max*.

- F) Researchers submitting manuscripts on new gene symbols are urged to furnish R. L. Bernard with seeds of the line carrying the reported gene. From 50 seeds to 300 gms of seed of each line are needed to maintain the genetic type collection. When these seeds are received, the genetic type number can be assigned and can then be reported by the author in a manuscript.

### Rules for Genetic Symbols

#### I) Gene Symbols

- a) A gene symbol shall consist of a base of one to three letters, to which may be appended subscripts and/or superscripts as described below.
- b) Genes that are allelic shall be symbolized with the same base letter(s) so that each gene locus will be designated by a characteristic symbol base.
- c) The first pair of genes reported for a gene locus shall be differentiated by capitalizing the first letter of the symbol for the dominant or partially dominant allele. (Example: *Ab*, *ab*. *Ab* is allelic and dominant to *ab*.) If genes are equivalent, codominant, or if dominance is not consistent, the capitalized symbol may be assigned at the author's discretion.
- d) When more than two alleles exist for a locus, the additional alleles or those symbolized subsequently to the pair first published shall be differentiated by adding one or two uncapitalized letters as a superscript to the base. (Example: *R*, *r<sup>m</sup>*, *r*.) This shall be the only use of superscripts. The base for the additional alleles is capitalized only when the gene is dominant or equivalent to the allele originally designated with a capitalized symbol. The superscript may be an abbreviation of a descriptive term. When allelism is discovered for a gene previously assigned a symbol, the previous symbol may be used as the superscript.
- e) Gene pairs with the same or similar effects (including duplicate, complementary or polymeric genes) should be designated with the same letter base differentiated by numerical subscripts, assigning 1, 2, 3, 4, etc., consecutively in the order of publication. (Example: The *y* series for chlorophyll deficiency.) This shall be the only use of subscripts. Letter subscripts should not be used. The subscript 1 is automatically a part of the first reported gene symbol for each base but may be omitted until the second symbol is assigned.
- f) Base letters may be chosen so as to indicate apparent relationships among traits by using common initial letters for all loci in a related group of traits. Examples are *P* for pubescence type, *R* for disease reaction (plus two initials of the pathogen to complete the base), and *L* for leaf shape.
- g) The distinction between traits that are to be symbolized with identical, similar, or with unrelated base letters is necessarily not clear cut. The decision for intermediate cases is at the discretion of the author but should be in accordance with previous practices for the particular type of trait. The following sections concern supplementary

symbols that may be used whenever desired as aids to presentation of genetic formulas.

- h) A dash may be used in place of a gene symbol to represent any allele at the indicated locus. The locus represented should be apparent from its position in the formula. (Example:  $A_{-}$  represents both  $AA$  and  $Aa$ .)
- i) A question mark may be used in place of a symbol when the gene is unknown or doubtful, or it may be used as a superscript to the base symbol for the same purpose. (Example:  $a^?$  indicates that the latter is an unknown allele at the  $A$  locus.)
- j) Plus symbols may be used in place of the assigned gene symbols of a designated standard homozygous strain when this will facilitate presenting genetic formulas. The standard strain may be any strain selected by the worker, as long as the strain being used and its genetic formula are made explicit.

## II) Linkage and Chromosome Symbols

- a) Linkage groups and the corresponding chromosomes shall be designated with Arabic numerals. Linkage shall be indicated in a genetic formula by preceding the linked genes with the linkage group number and listing the gene symbols in the order that they occur on the chromosome.
- b) Permanent symbols for chromosomal aberrations shall include a symbol denoting the type of aberration plus the chromosome number(s) involved. Specific aberrations involving the same chromosome(s) shall be differentiated by a letter as follows: The symbol *Tran* shall denote translocations. *Tran 1-2a* would represent the first case of reciprocal translocations between chromosomes 1 and 2, *Tran 1-2b* the second, etc. The symbol *Def* shall denote deficiencies, *Inv* inversions, and *Tri* primary trisomics. The first published deficiency in chromosome 1 shall be symbolized as *Def 1a*, the second as *Def 1b*, etc. The first published inversion in chromosome 1 shall be denoted as *Inv 1a*, etc. The first published primary trisomic shall be designated with the Arabic numeral that corresponds to its respective linkage group number.
- c) Temporary symbols for chromosomal aberrations are necessary, as it may be many years before they are located on their respective chromosomes. *Tran 1* would represent the first case of a published reciprocal translocation; *Tran 2*, the second case, etc. The first published deficiency shall be symbolized as *Def A*, the second as *Def B*, etc. The first published inversion shall be symbolized as *Inv A*, and second as *Inv B*, etc. The first published primary trisomic shall be designated as *Tri A*, the second as *Tri B*, etc. When appropriate genetic and/or cytological evidence is available, the temporary symbols should be replaced with permanent symbols, with the approval of the Soybean Genetics Committee.

## III) Cytoplasmic Factor Symbols

- a) Cytoplasmic factors shall be designated with one or more letters prefixed by *cyt-*. (Example: *cyt-G* indicates the cytoplasmic factor for maternal green cotyledons, *cyt-Y* indicates that for maternal yellow cotyledons.)

#### IV) Priority and Validity of Symbols

- a) A symbol shall be considered valid only when published in a recognized scientific journal, or when reported in the Soybean Genetics Newsletter, with conclusions adequately supported by data which establish the existence of the entity being symbolized. Publication should include an adequate description of the phenotype in biological terminology, including quantitative measurements wherever pertinent.
- b) In cases where different symbols have been assigned to the same factor, the symbol first published should be the accepted symbol, unless the original interpretation is shown to be incorrect, the symbol is not in accordance with these rules, or additional evidence shows that a change is necessary.

#### V) Rule Changes

- a) These rules may be revised or amended by a majority vote of the Soybean Genetics Committee





# VI. USDA SOYBEAN GERMPLASM REPORT

Approximately 147 strains were grown in 1980 and added to the USDA Soybean Germplasm Collection in 1981 from the following countries:

	<u>North</u>	<u>South</u>	<u>Total</u>
East Germany	51	--	51
Poland	23	--	23
Romania	13	--	13
China	16	14	30
South Korea	13	6	19
Indonesia	--	<u>11</u>	<u>11</u>
Total	116	31	147

The strains from Europe include some older European varieties not previously in the Collection. We hope to continue to complete our collection of European varieties, many of which are based on germplasm independently obtained from Asia many years ago. We are continuing to get Chinese varieties a few at a time through visitors or directly from research institutions in China, but still the government of China persists in preventing major shipments of germplasm from leaving the country.

Since the above strains were received, we have received an additional 389 strains which are being grown and will soon become available for seed requests. There were 98 from China, 1 each from Japan, Burma, Thailand, and India, and 289 from South Korea, consistently our best supplier of native Asian germplasm. We also have received 25 strains of wild soybeans from China which we are growing in 1982 to add to the Collection.

We have revised and updated (1900 to 1980) the draft of our "Soybean Germplasm Register" (information on source, origin, variety name, etc., on all strains in the Variety, FC, and PI Collections) and are looking toward a publication date this summer. We hope that this will be useful to all users of the Germplasm Collection. Many PI strains are named varieties in their country of origin, and American researchers should be using these names, along with the PI number, especially when publishing in journals that are read internationally.

At Urbana, we have divided the US and Canadian Variety Collection into two natural divisions. The first one, called "Germplasm Varieties," consists of all introduced varieties, those derived from introductions through mixture and outcrossing, and other old varieties developed before 1945. This corresponds closely to the type of named varieties included with the FC and PI strains in the southern collection at Stoneville, Miss. The second division, called "Public Varieties, Groups 000 to IV, US and Canada," consists of all varieties developed domestically since 1945. These are almost entirely derived from hybrids and the ancestral parents are in the Germplasm Varieties, FC, or PI Collection. Such derived varieties as this are often not included in germplasm collections of other crop species. However, we believe that this is an important service that we can easily provide along with the basic germplasm collection, and this is confirmed by the large number of requests for seeds and information that we fill. In view of the present activity in private soybean breeding in the US, we would like to extend this

service to include a third division, called "Private Varieties, Groups 000 to IV, US and Canada," and have sent a questionnaire concerning procedures for this to the private breeders. Response has been favorable; we expect to get this collection underway this year and will have further information to present on it at next February's workshop.

In 1981, a general evaluation of nearly 2800 introductions was completed with the assistance of Dr. J. H. Orf and Dr. J. W. Lambert. This evaluation began with PI 273.483A and included all introductions in Maturity Group IV or earlier through PI 427.107C. We are anticipating that the results will be available before the end of the year.

This spring, again with the cooperation of Dr. Orf, we will begin a similar evaluation of over 1800 accessions added to the collection since 1980. Nearly 1700 of these strains are from the large and valuable soybean collection of the Vavilov Institute of Plant Industry in Leningrad, U.S.S.R. Almost 900 of these accessions originated in China and another 300 were from areas of the U.S.S.R. which border northeast China. This exchange of germ-plasm added to our collection more accessions from China than have been added from all other collection trips and exchanges combined! An additional 250 strains were collected in 18 different areas of the U.S.S.R. from a wide range of both latitude and longitude. The 1982-83 evaluation will also include strains from South Korea and Eastern Europe.

100 R. L. Bernard

R. L. Nelson

## VII. RESEARCH NOTES

Brazil

EMPRESA BRASILEIRA DE PESQUISA AGROPECUARIA  
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X) <sup>245</sup> Reaction of soybean cultivars to Temik (aldicarb) and inheritance of the reaction<sup>[12]</sup>.

Temik (aldicarb) is a systemic <sup>1</sup>insecticide not commonly used in soybeans. Soybean researchers in general have problems to control spider mites in greenhouses. Temik proved to be an excellent product to control these pests. We started using the product in 1974 and noted that, with usual rates, it controls the mites; some soybean cultivars show symptoms in the leaves very similar to the ones caused by soybean mosaic virus, while others remain free of symptoms. The symptoms appear on the newly formed leaves and should not be confused with the burning of the border of the leaves caused by excessive amounts of the product. The cultivars that showed the reaction we called sensitive while the others that remained free of symptoms we called tolerant.

The reaction to Temik was used in some cases by us to identify mixtures in seed lots. Because of its use as a possible test to identify cultivars, we classified, in 1977 and 1978, the most common cultivars used in Brazil as well as some cultivars that were of interest for breeding purposes (Table 1 shows the reaction of these cultivars).

In 1977, we made three crosses involving two sensitive cultivars ('Paraná' and 'Bossier') and one tolerant ('Davis'). In 1978, we made a fourth cross involving the cultivars 'IAS-5' (sensitive), and 'Davis'. For all crosses we used single plant selections. The single plant selection of Paraná used in the crosses was a sensitive one.

The cross between two sensitive cultivars (Bossier x Paraná) showed in the F<sub>2</sub> all 126 plants sensitive.

The F<sub>2</sub> generation of the crosses between tolerant and sensitive parents segregated in a 3:1 ratio with tolerant dominant (Table 2).

Table 1. Reaction of soybean cultivars to Temik (aldicarb)

Tolerant	Sensitive	Tolerant	Sensitive
BR-2	Bacatete	Davis	Cutler 71
BR-3	Bossier	Florida	Forrest
Bragg	BR-1	Hardee	Hampton
Campos Gerais	Cajeme	Hood	Hill
Co-136	Clark 63	IAC-1	IAIREEN
NS-4	CNS	IAC-4	IAC-3
Cobb	Columbus	IAC-7	IAC-5
IAS-4	IAC-6	Pelicano	Oriente
Majos	IAS-1	Pérola	Paraná*
Mitchell	IAS-2	Planalto	Prata
Missões	IAS-5	Sant'Ana	Sulina

Table 1. *Continued*

Tolerant	Sensitive	Tolerant	Sensitive
Mineira	Lancer	São Luiz	Viçosa
Pampeira	Lee 68	UFV-2	UFV-1

\*Shows sensitive and tolerant plants.

Table 2. Segregation of the reaction to Temik (aldicarb) in the F<sub>2</sub> generation of three crosses

Cross	Number of plants		Total	X <sup>2</sup> value	P value (3:1)
	Tolerant	Sensitive			
Davis x Bossier	57	17	74	0.162	0.75 - 0.50
Paraná x Davis	79	17	96	2.722	0.10 - 0.05
IAS-5 x Davis (1)	84	20	114	0.150	0.75 - 0.50
IAS-5 x Davis (2)	171	61	232	0.207	0.75 - 0.50

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#### 2) Calico mosaic of soybean: Sources of resistance and inheritance of reaction [ ]

Calico mosaic of soybean is caused by the alfalfa mosaic virus (AMV) (Almeida et al., 1981). Soybean plants infected with AMV were first reported in the USA by Allington et al. (1969), in soybean fields close to alfalfa fields. The presence of AMV in Brazil was reported by Costa et al. (1980), who demonstrated in their study that soybean was a good differential host for the virus.

Soybean plants naturally infected with AMV were found in Londrina, PR, in 1981, by Almeida et al. (1981).

The disease caused by the AMV is apparently of minor importance in the areas of the world where it has been reported. It is expected that this disease will be of minor importance in Brazil also, but two aspects favor the disease development: the AMV is seed transmitted (Costa et al., 1981) and it is easily transmitted from plant to plant of soybean by aphids.

In general, resistance is the best way to control diseases; therefore, we started screening germplasm for resistance. The screening started with the most common soybean cultivars in Brazil. Two among the first 26 tested proved to be resistant ('Pérola' and 'Planalto') (Table 1).

Both cultivars have 'Hood' as a common ancestor. Therefore, this cultivar was tested with AMV, and 119 plants were resistant and 129 plants were susceptible. Therefore, Hood is the possible source for the resistance of Pérola and Planalto. The other parents are being tested to confirm this hypothesis.

We used remnant  $F_2$  seeds from two crosses having Pérola as one parent, for preliminary inheritance studies (Table 2).

All plants of Pérola showed no symptoms and all plants of 'Co-136' and 'Dourados' showed typical calico mosaic symptoms. In the  $F_2$  generation, the resistant and susceptible plants showed proportions that fitted a 3:1 ratio. All chi-square values for tests of goodness of fit, within crosses and pooled over crosses, were acceptable.

Other crosses are being made to get better information on the heterozygous reaction.

Table 1. Reaction of 26 soybean cultivars upon inoculation with AMV

Variety name	Disease reaction <sup>a</sup>	Variety name	Disease reaction <sup>a</sup>
Andrews	S	IAC-2	S
Bragg	S	IAC-3	S
Bienville	S	IAC-4	S
Bossier	S	IAC-5	S
BR-1	S	Mineira	S
BR-2	S	Paraná	S
BR-3	S	Pérola	<u>R</u>
Campos Gerais	S	Planalto	<u>R</u>
Co-136	S	Santana	S
Davis	S	São Luiz	S
Dourados	S	Santa Rosa	S
Flórida	S	UFV-1	S
IAC-1	S	Viçoja	S

<sup>a</sup>R = resistant and S = susceptible.

Table 2. Segregation of the reaction to AMV in the F<sub>2</sub> generation of two crosses and the reaction of the parents

Cultivar or cross	Generation	— Number of plants — Resistant Susceptible	Total	$\chi^2$ Value	P Value (3:1)
Pérola	P1	19 0	19		
Dourados	P2	0 19	19		
Coker 136	P3	0 19	19		
Pérola x Dourados (1)	F <sub>1</sub>	(120.0) <sup>+</sup> 50	160	3.333	0.10 - 0.05
Pérola x Dourados (2)	F <sub>2</sub>	(90.0) 32	120	0.178	0.75 - 0.50
Pérola x Dourados (3)	F <sub>2</sub>	(109.5) 40	146	0.448	0.75 - 0.50
Coker 136 x Pérola	F <sub>2</sub>	(131.25) 42	175	0.093	0.90 - 0.75

<sup>+</sup>Expected numbers in parentheses.



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- Allington, W. B., E. L. Moorhead and R. Staples. 1969. Alfalfa mosaic virus in soybeans. *Phytopathology* 50:627 (Abstract).
- Almeida, A. M. R., A. Bianchini, A. S. Costa and J. Vega. 1981. Mosaico cálico: nova virose da soja no Brasil. *Fitopatol. Bras.* (in press).
- Costa, A. S., J. Vega and G. A. Groppó. 1980. Ocorrência do vírus do mosaico da alfalfa em São Paulo. In: *Resumos do II Congresso Paulista de Fitopatologia*, p. 52.
- Costa, A. S., G. A. Groppó and J. Vega. 1981. Transmissão do vírus do mosaico da alfalfa através da semente de soja. In: *Anais do II Seminário Nacional de Pesquisa de Soja*. Brasília (in Press).

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h) <sup>245</sup> Soybean linkage and crossover tests,

A previous test (Buzzell, 1977) indicated a possible linkage ( $39.8 \pm 3.0\%$ ) between  $Fg_1$  and  $Dt_1$ . An additional test with  $F_2$  plants of a cross of two 'Harosoy' isolines ( $fg_1 dt_1 \times Fg_1 Dt_1$ ) indicates that the two genes are independent (Table 1).

In reporting on the genetics of flavonol classes 9T to 16T, Buzzell and Buttery (1974) indicated that the 9t to 16t classes which involve  $fg_4 t$  had not been observed. A close linkage between  $Fg_4$  and  $T$  was apparent. In the early generation of a cross between OX 936 ( $fg_4 T$ ) and 'Beeson' ( $Fg_4 t$ ), the crossover type was not found among gray pubescent plants. Without a crossover to break the linkage, the following segregations were expected from 8T ( $T fg_4/t Fg_4$ ) plants:

Class	Linked genotype		Ratio	Segregation without crossover
16T	$T fg_4$	$T fg_4$	1	All brown
8T	$T fg_4$	$t Fg_4$	2	Brown and gray
8t	$t fg_4$	$t fg_4$	1	All gray

On this basis, the following procedure was used. In rows segregating for brown and gray pubescence, the gray plants were discarded and the brown plants were sampled for thin layer chromatography (TLC) and harvested for seed. The plants were separated into 16T and 8T classes and progeny were grown in rows. The 16T rows were checked for pubescence color; when no segregation was observed, the rows were discarded and the cycle was repeated with the 8T rows. In a population of 299 rows derived from  $F_8$  16T plants, several rows were segregating brown and gray pubescence. TLC of gray plants showed that they were 16t.

Table 1. Soybean  $F_2$  linkage test

Genes		a	b	c	d	Sum	R%	SE
$Fg_1 fg_1$	$Dt_1 dt_1$	165	58	49	13	285	54.0	4.6

References

- Buzzell, R. I. 1977. Soybean linkage tests. Soybean Genet. News1. 4:12-13.
- Buzzell, R. I. and B. R. Buttery. 1974. Flavonol glycoside genes in soybeans. Can. J. Genet. Cytol. 16:897-899.

180 R. I. Buzzell  
R. D. Walker

2) <sup>245</sup> Inheritance of presence/absence of flavonoid compounds in soybean seed coats.

In soybean plants carrying the gene *T* and having black or brown pigmentation of seed coats, there are numerous compounds that can be detected by thin layer chromatography (TLC). There are four spots (A, B, C and D) that appear to be related; they are yellow-orange under visible light and they fluoresce yellow-orange (duller than quercetin) under UV light after spraying with flavone reagent. Phenotypic positions of A and D on 2-way plates are given in Table 1. A is present in all material tested, but D varies in presence/absence (B and C also vary but have not been studied). Twenty-nine cultivars, two lines, 15 genetic types, and 78 plant introductions were evaluated for leaf flavonol class and for seed coat compounds. The TLC technique followed that of Buttery and Buzzell (1973). The presence of D was associated with *Fg*<sub>2</sub> in all of the entries tested (Table 2). Segregations support the association (Table 3). Either there is a gene closely linked with *Fg*<sub>2</sub> or else there is a "pleiotropic effect" of *Fg*<sub>2</sub> on D. *Fg*<sub>2</sub> is a flavonol glycoside gene for adding rhamnose with an  $\alpha(1-6)$  linkage (Buttery and Buzzell, 1975). If *Fg*<sub>2</sub> is involved, the D could be a rhamnose glycoside of a flavonoid compound.

There is another group of compounds that appear to be related to the A-D compounds. They fluoresce a dull orange. We have labeled them *theta* ( $\theta$ ) spots. When present, they occur just to the right of A, B, C and D on the TLC plate. Segregation for A $\theta$  indicates that a dominant gene is involved in its presence (Table 4). When this gene and *Fg*<sub>2</sub> are present, D $\theta$  occurs.

Table 1. Phenotypic positions of two seed coat compounds on 2-way TLC cellulose plates

Run	Spot A		Spot D	
	Rear	Front	Rear	Front
First <sup>b</sup>	21	39	50	58
Second <sup>c</sup>	95	104	93	100

<sup>a</sup>From point of spotting, with runs 140 mm; averages of measurements on 10 cellulose plates.

<sup>b</sup>2% Formic acid.

<sup>c</sup>Amyl alcohol (20): acetic acid (12): water (10).

Table 2. Numbers of black- and brown-pigmented entries by leaf-flavonol classes that have either  $Fg_2$  or  $fg_2$  and either presence or absence of compound D in seed coats

Seed coat color	$Fg_2$ present and Spot D present				
	<u>1T</u>	<u>2T</u>	<u>4T</u>	<u>6T</u>	<u>14T</u>
Black	2	7	11	38	0
Brown	0	4	9	18	1

	$Fg_2$ absent and Spot D absent			
	<u>3T</u>	<u>5T</u>	<u>7T</u>	<u>8T</u>
Black	0	1	2	2
Brown	0	0	29	0

Table 3. Segregation of  $Fg_2/fg_2$  and numbers of plants with and without spot D in the  $F_2$  of Lincoln-i x Beeson i

Leaf flavonol class	Spot D	
	Present	Absent
6T ( $Fg_2$ )	50	0
8T ( $fg_2$ )	0	17

Table 4. Segregation for a flavonoid compound of soybean seed coats

T31 x OX936	A0		3:1 test	
	Present	Absent	Chi-square	P
$F_2$ -91 (A0 absent) $F_3$	0	29	--	--
$F_2$ -93 (A0 present) $F_3$	31	14	0.60	0.50-0.40

#### References

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- Buttery, B. R. and R. I. Buzzell. 1975. Soybean flavonol glycosides: Identification and biochemical genetics. Can. J. Bot. 53:219-224.

100 R. I. Buzzell  
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### 3) <sup>247</sup> Genetics of black pigmentation of soybean seed coats/hila,

Gene  $T$  is involved in black pigmentation and  $W_1$  in the presence of  $t$  is involved in imperfect black pigmentation (Bernard and Weiss, 1973).  $T$  is a phenolase gene resulting in quercetin formation and brown pubescence (Buttery and Buzzell, 1973);  $W_1$  is a flower- and hypocotyl-color gene (Hartwig and Hinson, 1962). Thin layer chromatography shows two major 'spots' involved in black pigmentation of soybean seed coats. Phenotypic descriptions are given in Table 1. Evaluation of the seed coats of 23 cultivars, 2 lines, 11 genetic types, and 36 plant introductions having black pigmentation indicated that spot #1 is associated with the presence of  $W_1$  and that the presence of spot #2 is associated with  $T$  (Table 2). This association is supported by the segregations listed in Table 3. Preliminary analyses at Harrow suggest that spot #1 is a delphinidin and spot #2 a cyanidin glycoside. Yoshikura and Hamaguchi (1969) have identified delphinidin 3-monoglucoside and cyanidin 3-monoglucoside as anthocyanins of black soybean.

Other genes were tested for possible effects upon seed coat anthocyanins (Table 4). The genes  $w_3$  and  $w_4$  (Hartwig and Hinson, 1962) did not affect the presence of spots #1 and #2. The gene  $w_m$  (Buzzell et al., 1977) did not affect the presence of spot #2 (the  $W_1 w_m$  combination was not tested). The gene  $td$  (Bernard, 1975) did not affect the presence of spots #1 and #2. In addition, spot A (Buzzell and Buttery, 1982) was not affected by any of these genes.

Table 1. Phenotypic description of anthocyanins of soybean seed coats on 2-way TLC plates

Run <sup>a</sup>	Spot #1 <sup>c</sup>		Spot #2 <sup>d</sup>	
	Rear	Front	Rear	Front
mm <sup>b</sup>				
<u>Imperfect black seed - Purple flowers - Gray pubescence</u>				
First	14	20	--	--
Second	46	54	--	--
<u>Black seed - White flowers - Brown pubescence</u>				
First	--	--	20	45
Second	--	--	68	87
<u>Black seed - Purple flowers - Brown pubescence</u>				
First	15	37	20	49
Second	45	53	71	86

<sup>a</sup>First -- 2% formic acid; Second -- amyl alcohol (20): acetic acid (12): water (10).

<sup>b</sup>From pointing of spotting, with 140-mm runs; averages of measurements of each type on 5 cellulose plates.

<sup>c</sup>Unsprayed: red-purple in visible light. Sprayed with flavone reagent: blue in visible light, fluoresces red-purple.

<sup>d</sup>Unsprayed: red-purple in visible light. Sprayed with flavone reagent: varies in intensity within the spot from blue to violet-blue in visible light, fluoresces red-purple.



Table 2. Association of anthocyanins of soybean seed coats with genes  $W_1$  and  $T$ 

Seedcoat color	No. of entries	Flower color	Spot #1	Pubescence color	Spot #2
Black	30	Purple ( $W_1$ )	Present	Brown ( $T$ )	Present
Black	2	Dilute purple ( $W_1W_3W_4$ )	Present	Brown ( $T$ )	Present
Black	35	White ( $w_1$ )	Absent <sup>a</sup>	Brown ( $T$ )	Present
Imperfect-black	4	Purple ( $W_1$ )	Present	Grey ( $t$ )	Absent <sup>a</sup>

<sup>a</sup>Not detectable with technique used but may be present in trace or low amounts.

Table 3. Segregations for  $W_1/w_1$  and  $T/t$  showing association with anthocyanins for black pigmentation of soybean seed coats

Cross	Generation	Gene	Spot #1		Spot #2	
			Present	Absent <sup>a</sup>	Present	Absent <sup>a</sup>
T31 ( $T$ ) x OX 936 ( $T$ )	$F_3$	$W_1$	53	0	53	0
		$w_1$	0	18	18	0
Lincoln- $i$ x Beeson- $i$	$F_2$	$W_1$	70	0	53	0
		$w_1$	0	24	15	0
		$T$	53	0	68	0
		$t$	17	0	0	26

<sup>a</sup>Not detectable with technique used but may be present in trace or low amounts.

Table 4. Tests for effects of other genes on seed coat compounds

		<u>Spot A</u>	<u>Spot #1</u>	<u>Spot #2</u>
$w_3$ and $w_4$				
L67-3469 <sup>a</sup>	$T W_1 w_3 W_4 i R$	Present	Present	Present
OX 326 <sup>b</sup>	$T W_1 W_3 w_4 i R$	Present	Present	Present
OX 327 <sup>c</sup>	$T W_1 w_3 w_4 i R$	Present	Present	Present
$w^m$				
OX 694 <sup>d</sup>	$T w_1 w^m i R$	Present	Absent*	Present
$td$				
Cloud	$T td w_1$	Present	Absent*	Present
T 69	$T td w_1$	Present	Absent*	Present
Kingwa	$T td W_1$	Present	Present	Present
Sooty	$T td W_1$	Present	Present	Present

<sup>a</sup>Clark isolate from Dr. R. L. Bernard.

<sup>b</sup>From L70-4422 ( $w_1 w_3 w_4 i^i$ ) x L67-3469.

<sup>c</sup>From L68-1774 ( $w_1 w_3 w_4 i^i$ ) x L67-3469.

<sup>d</sup>From OX 281 ( $w_1 w^m I t r$ ) x Harman ( $w_1 w^m i TR$ ).

\*Not detectable with technique used but may be present in trace or low amounts.

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4) <sup>245</sup> Soybean emergence in clay soil and tolerance to phytophthora rot [77],

In 1979 at the Woodslee Soil Substation, 19 F<sub>2</sub> populations were planted May 23 in single-row plots, 100 seeds per row, with 12 replications. These populations consisted of 'Williams' x 'Wells', and Williams and Wells each by 'Adams', 'Beeson', 'Bonus', 'Calland', 'Corsoy', 'Cutler 71', 'Hark', 'Kanrich' and 'Prize'. The field had been in continuous soybeans since 1973 and is known to be infested predominantly with race 7 of *Phytophthora megasperma* f. sp. *glycinea* (Pmg).

Emergence was slow as a result of heavy rainfall (7 cm in the week after planting followed by 12 cm during the next three weeks) and soil compaction/crusting. Emergence counts were taken June 21. Diseased plants were recorded and removed from the plots biweekly until maturity.

The amount and occurrence of phytophthora rot was small and variable, thus comparisons of each cross could not be made; and, although some rows of parents were planted, results are not presented because seed of similar quality/viability was not available for all of them. Each of the varieties is susceptible to race 7 of Pmg with hypocotyl inoculation.

Wells is moderately susceptible to Pmg in the field (Buzzell and Anderson, 1982); Williams is moderately tolerant (Schmitthenner and Walker, 1979). Averages were run over the nine progenies for Wells and for Williams (Table 1). Williams progenies had higher percentage of emergence and less plant loss than did Wells progenies. The Williams x Wells cross was intermediate, suggesting polygenic inheritance.

#### Acknowledgments

The F<sub>2</sub> seed lots were provided by R. L. Bernard and R. L. Nelson, University of Illinois. The Ontario Soya Bean Growers' Marketing Board provided help in making plant counts.

Table 1. Seedling emergence in clay soil and plant loss from phytophthora rot

	Emergence (%)	Plant loss (%)
Williams progenies*	46 b	2 a
Williams x Wells	41 ab	6 b
Wells progenies*	38 a	8 b
SE	1.8	1.0
C.V. %	15	66

\*Average of nine crosses.

a-b Means in columns followed by the same letter do not differ significantly ( $P = 0.05$ ) by Duncan's multiple range test.

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## 5) Greenhouse determination of soybean tolerance to phytophthora rot [1,2].

One method of determining field tolerance of soybeans to *Phytophthora megasperma* f. sp. *glycinea* (Hildeb.) Kaun & Erwin (Pmg) is to determine the percent of plant loss from emergence to maturity for [cultivars] grown in an infested field under conditions favorable for the disease (Buzzell and Anderson, 1982). Drawing upon the field results and the results of two unpublished greenhouse experiments (given below), a greenhouse technique similar to the field test was developed.

1962 Experiment. Cores of soil were obtained from a field known to be infested with Pmg race 1, the field having been used in a previous study (Fulton et al., 1961). In the fall of 1961, each of 40 pieces of aluminum pipe (17 cm diameter by 24 cm) was pressed 20 cm into the ground using a hydraulic jack, dug up containing soil, and moved to a greenhouse. Each was set on a tin plate as a "pot," with pots making up a five-replicate test. Allowing

for volume increases, the soil was tilled with a trowel to a sufficient depth to allow for four planting treatments of 0, 38, 76, and 114 mm of worked soil between the seeds and the untilled soil layer for each of 25 and 50 mm seed depths of planting. Twenty-five seeds of 'Harosoy' were planted per pot January 8-9, 1962. The number of plants with phytophthora rot were recorded from January 28 to March 22.

The depth of seed placement at 25 and 50 mm had no effect on the incidence of phytophthora rot. There was no interaction between depth of seed and distance of seed from the untilled soil layer. The average loss (over depths) of Harosoy plants killed by the disease was 36, 30, 12, and 13% for the 0, 38, 76, and 114 mm distances of seed to untilled layer; the L.S.D. 0.05 was 15%. It was concluded that seed placement at or near the untilled layer increased the amount of phytophthora rot.

1974 Experiment. Soil was collected from around dead soybeans in 31 fields. The soils were placed in 30-cm-diameter fibre pots in a greenhouse. Repetitive plantings of 'Amsoy', Harosoy, and 'OX20-8' soybeans as a trap crop were made in each soil for 4 to 6 times. The number of emerged seedlings and the number of killed seedlings were recorded. Pmg (races 3, 4, and/or 6) was isolated from some of the killed seedlings (Buzzell et al., 1977); Amsoy, Harosoy and OX20-8 are susceptible to each of these races. Each of the cultivars differed significantly ( $P = 0.05$ ) from the others in the percentage of plants killed by phytophthora rot: Amsoy 4%, Harosoy 10%, and OX20-8 26%.

Tolerance Experiment. Clay soil was obtained from a field being used for phytophthora tolerance tests (Buzzell and Anderson, 1982). Forty-eight fibre pots (20 cm diameter, 23 cm tall) were filled with 18 cm of soil after placing glass wool in the drainage holes. The soil was tamped into the pots with a small weight, then watered. Three plantings of the extremely susceptible OX20-8 were made from January to April, 1978. Interspersed with these plantings were two plantings of a 3-replicate test of 15 cultivars ('Altoona', 'Amsoy 71', 'Dawn', 'Harcor', 'Harlon', 'Harosoy 63', 'Harwood', 'Maple Arrow', 'Nairn', OX20-8, 'Starbuck', 'Steele', 'Viking', 'Wells', 'XK505') covering a range of field tolerance along with 'Toyosuzu' as a resistant check. Twenty-five seeds per pot were placed onto the clay surface and covered with about 3 cm of builder's sand. The pots were watered as required, especially as seedlings were emerging to break up crusted sand and to wash the sand back down around the base of the seedlings. Counts were made of emerged seedlings and of live plants 18 to 26 days from planting. For a few days prior to the final reading, the pots were allowed to dry out sufficiently to place the seedlings under some moisture stress. The sand became dry which meant that if there were seedlings sufficiently diseased to not be rooted into the soil, their death was hastened. The same soil was used in each planting, without being tilled. The seedlings were cut off and removed without disturbing the soil. The dry sand was dumped into tubs and re-used in the next planting.

There was no plant loss in the resistant check. The two later plantings of OX20-8 had more plant loss (60 and 63%) than the first planting (45%), indicating a possible buildup in inoculum. Likewise the second planting of the cultivar test averaged higher plant loss (27%) than the first (14%). Averaged over the two plantings there were significant ( $P = 0.01$ ) cultivar differences in plant loss; the C.V. was 12%. There was a positive correlation between the greenhouse plant losses and unpublished field plant losses from 1974 and 1975 ( $r = 0.75^{**}$ ) and field plant losses (Buzzell and Anderson, 1982) from 1977 and 1978 ( $r = 0.79^{**}$ ).



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101 C. G. Mortimore - retired

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1) <sup>245</sup> N-23-A, a new promising variety of soybean for Chotanagpur,

Soybean is a unique dual-purpose leguminous crop containing high quantity of protein (40%) along with sufficient (20%) extractable edible oil. Its nutritive value is very highly suited for versatile industrial uses. Its increasing industrial exploitation also has led to the manufacture of a large number of antibiotics in our country. According to Singh (1978) it is the cheapest, richest and best source of protein and fat. Its seed yield is highest among pulses and oilseed crops and production is easiest and most economical.

Soybean is a highly lime-responsive crop and can be grown economically in the acidic soils of the uplands of Chotanagpur (Bihar) with the application of lime (Mandal, 1962). Well-drained upland soils of this region have been found to be ideally suited for soybean cultivation.

In the past, 'Punjab-1' was found to be the most promising soybean variety for this region (Bhargava and Rao, 1972), but with the intensification of research work on this crop in the state of Bihar under I.C.A.R. Co-ordinated Research Project, 'Bragg' and 'Alankar' varieties were recommended to replace Pb-1 for the uplands of the plateau region of Bihar. This region has special advantage over other regions in India in that soybeans are found to be free from yellow mosaic virus which is a serious disease of this crop in India.

During the subsequent years under the coordinated varietal trials conducted at Ranchi Agricultural College, Kanke variety 'N-23-A' has shown better promise than Bragg and Alankar during the last three years. This variety is evolved from 'Sepaya Black' out of the progeny of a white-seeded natural mutant plant detected in the field of a black-seeded variety.

Three-years yield data and combined analysis of 14 varieties included in the trials are presented in Table 1. The results indicate that N-23-A is superior to Alankar in seed yield and also is consistent in performance. Alankar was highest yielding variety of this region. N-23-A has also shown promise in seed yield over other entries in minikit trials conducted in farmers' fields and government farms (Table 2). In all-India coordinated trial (1980), N-23-A gave highest seed yield in Delhi, Ranchi and Palampur.

N-23-A (Birsa Soy.I) has white flowers, golden brown pods, dark green foliage with tawny pubescence, medium sized (47 cm) erect stem, height of the plant is nearly twice the length of the main stem because of its upright standing position of leaves, a unique favorable characteristic for efficient utilization of solar energy for photosynthesis. Seeds are bold shining black with dull white hilum, oval in shape; 100-seed weight is 12.5 gms. There is 41.7% protein and 20.4% oil in the seeds. Their germination quality is much better than Alankar.

N-23-A is found resistant to bacterial pustule and rust diseases. It was also found to be resistant to yellow mosaic, phytophthora root rot and downy mildew under prevailing climatic conditions of Ranchi.

Table 1. Seed yield (kg/ha)

Sl. no.	Varieties	Seed yield (kg/ha)			Mean	Rank
		1978-79	1979-80	1980-81		
1.	N 19	2407	1917	2625	2316	II
2.	I15A	2259	1667	2264	2063	IX
3.	I8	1907	1694	2222	1941	X
4.	NA-2	2573	2028	2208	2269	III
5.	I-14B	2203	1986	2195	2128	VII
6.	N-3	2518	1775	2167	2153	VI
7.	N-7	2055	1897	2153	1935	XI
8.	N-23-A	2870	2195	2097	2387	I
9.	N-22	2425	2195	2097	2239	IV
10.	I-13B	2073	1736	1958	1922	XII
11.	I-2	2475	1944	1931	2100	VIII
12.	Alankar	2851	1889	1819	2186	V
13.	S. Black	2018	903	1681	1534	XIV
14.	I-6	2370	1542	1431	1781	XIII
SEm + (kg/ha)		209	130	469	176	
C.D. at 5% (kg/ha)		430	358	N.S.	494	
C.V. %		10.87	13.98	22.81	14.77	

Table 2. Seed yield (kg/ha) in minikit trials

Sl. no.	Varieties	Cultivator's field '79-80			Mean	1980-81	Mean
		Ormanjhi	Nagri	Sukurhuttu		Govt. farm	
1.	NA-2	13.95	5.19	9.42	9.92	25.50	17.71
2.	N-23-A	14.51	8.56	15.82	13.98	24.80	19.39
3.	Alankar	12.08	4.88	12.98	9.98	22.50	16.24
4.	N-22	13.88	5.87	11.47	10.07	21.50	15.63

Considering its superiority in yield potential and consistency in performance in fluctuating weather conditions of this tract over Alankar, N-23-A (Birsa Soy.I) will be recommended shortly for release by the Research Council of Birsa Agricultural University, Bihar for cultivation in Chotanagpur.

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### 245 Effect of growth regulators Cycocel (CCC), Regim-8 (TIBA) and Ethrel (CEPA) on soybean crop

Summary: Soybean variety 'Punjab-1' was sown in the research area of the Ranchi Agricultural College, Kanke, Ranchi, for studying the effect of the three growth regulators, viz.: Cycocel (CCC), Regim-8 (TIBA), and Ethrel (CEPA), on the crop yield. All the three chemicals significantly out-yielded the control. The maximum yield (2914 kg/ha), however, was obtained as the result of one foliar spray of Cycocel (750 cc/ha) 10 days after flowering, which was closely followed by Regim-8 at 100 cc/ha (2790 kg/ha). The increase in yield was 39.7% with Cycocel and 33.7% with Regim-8 as compared to control. All three growth-regulating chemicals also caused marked reduction in dry weight of vegetative plant parts with concomitant increase in dry weight of reproductive plant parts, which resulted in increased yield of soybean.

Introduction: Cycocel (2, chloro-ethyltrimethyl ammonium chloride), Regim-8 (Triiodobenzoic acid) and Ethrel (2, chloro-ethyl phosphoric acid) are plant-growth regulators that produce varied responses in a wide variety of crop plants. The primary action of CCC is to shorten the length of stem internodes and, thereby, reduce plant dry weight (Humphries and Dyson, 1967; Gunasena and Harris, 1969; Gunasena, 1970a, 1970b). This effect is reported to be due to inhibition of gibberellic biosynthesis (Reid and Carr, 1967). CCC-treated plants, after recovery from moisture stress, yielded more in comparison to untreated plants (Gill and Singh, 1978). Regim-8 has been found to produce a vertical orientation of leaves and triangular-shaped row canopy and thereby promote more efficient utilization of solar energy because of less self-shading by leaves in soybean crop (Greer and Anderson, 1965; Wax and Pendleton, 1968). The dry weight of vegetative plant parts was reduced, while reproductive plant parts were increased due to the above effect (Rao and Agrawal, 1981, unpublished). Ethrel, on application to plant, releases ethylene gas which is a broad-spectrum physiological agent in plant metabolism. With the above background knowledge of these growth

regulators, a trial was conducted to quantify the benefits to soybean crop in terms of extra yield.

Materials and Methods: The experiment was conducted for three consecutive years, 1978 through 1980, at Ranchi Agricultural College Farm, Kanke, Ranchi. The treatments were, Cycocel at 150, 200 and 750 cc/ha, Regim-8 at 50, 75 and 100 cc/ha. and Ethrel at 1500, 2000 and 2500 cc/ha. All the chemicals were sprayed at 10 days after first flowering. There was one control treatment with no spraying. The soybean variety Punjab-1 was sown at 45-cm row-to-row distance and plants were thinned at 5 cm apart in a randomized block design. The normal manurial schedule 80:60:40 kg NPK/ha was followed. Standard plant protection measures were adopted as and when required.

Results and Discussion: The data of mean yields and mean dry weight of vegetative and reproductive plant parts are furnished in Table 1, which will show that in each of the three years of trial, all the treatments significantly out-yielded the control. During the first year, the treatment yields varied from 2933 to 3413 as against 2447 kgs in the control. In the second and third years, the treatment yields ranged from 2240 to 2925 and 1946 to 2404 kgs as against 2148 and 1692 kgs in control, respectively. In all three years, the highest yields were recorded by Cycocel at 750 cc/ha. But in the first year, the Cycocel at 200 cc/ha and all the three doses of Regim-8 and of Ethrel were at par with this treatment; in the second year only Regim-8 at 100 cc/ha and Ethrel at 2500 cc/ha were at par while in the third year, both the lower doses of Cycocel (150 and 200 cc/ha), all the three doses of Regim-8 and Ethrel at 2500 cc/ha were at par with this treatment. In the combined analysis of the three years' data, this treatment (Cycocel 750 cc/ha) significantly out-yielded all the treatments including control, except Regim-8 at 100 cc/ha. Thus, Cycocel at 750 cc/ha proved the best treatment in increasing the yield of soybean closely followed by Regim-8 at 100 cc/ha. The increases in mean yields over the control due to the application of Cycocel at 750 cc/ha were 40.6, 36.2, 42.1% in 1978, 1979 and 1980, respectively and 39.7% over the three years' average yield, the corresponding increases due to Regim-8 at 100 cc/ha being 37.3, 29.3, 33.5 and 33.7%.

An examination of the data of mean dry weights of vegetative and reproductive plant parts will show that the application of those three growth regulators in each concentration had markedly reduced dry weights of vegetative plant parts and increased those of the reproductive plant parts as compared to the control, the reduction percentage varying from 26.3 to 61.3 and the percentage increase ranged between 14.3 and 85.7 in the different treatments. The highest reduction and increase was recorded by Cycocel at 750 cc/ha followed by Regim-8 at 100 cc/ha. These results are in agreement with those of Humphries and Dyson (1967).

#### Acknowledgment

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Table 1. Mean grain yields of vegetative and reproductive parts

Treatment	Mean grain yield in kg/ha			
	1978	1979	1980	Combined
Cycocel-150 cc/ha	2933 b	2481 cd	2084 ab	2499 de
Cycocel-200 cc/ha	3147 ab	2574 be	2176 ab	2633 bcd
Cycocel-750 cc/ha	3413 a	2925 a	2404 a	2914 a
Regim-8-50 cc/ha	3333 a	2611 bc	2156 ab	2700 bc
Regim-8-75 cc/ha	3173 ab	2555 bc	2164 ab	2631 bcd
Regim-8-100 cc/ha	3333 a	2777 ab	2259 ab	2790 ab
Ethrel-1500 cc/ha	3147 ab	2240 de	1946 bc	2444 e
Ethrel-2000 cc/ha	3120 ab	2555 bc	2020 b	2565 cde
Ethrel-2500 cc/ha	3227 ab	2759 ab	2211 ab	2732 bc
Control	2427 c	2148 e	1692 c	2086 f
S. Em	100.5	52.3	108.5	57.7
C.D. at 5%	299.0	269.0	323.0	172.0
C.V.	5.6	6.1	8.9	3.8

N.B.: Figs. with different letters differ significantly at  $P \leq 0.05$ .

Table 2. Mean dry weight of vegetative and reproductive parts

Treatment	Mean dry weight in 'g' at 85 DAS*							
	Vegetative plant parts				Reproductive plant parts			
	1978	1979	1980	Mean	1978	1979	1980	Mean
Cycocel-150 cc/ha	9.9	9.7	9.2	9.6	3.2	2.7	3.8	3.2
Cycocel-200 cc/ha	9.8	9.5	8.3	9.2	4.6	3.4	3.8	3.9
Cycocel-750 cc/ha	8.4	9.0	7.9	8.4	5.5	5.3	4.8	5.2
Regim-8-50 cc/ha	15.0	20.5	10.6	15.3	3.4	3.0	3.9	3.4
Regim-8-75 cc/ha	14.7	19.9	9.6	14.7	5.5	4.8	2.2	4.2
Regim-8-100 cc/ha	12.8	19.8	9.2	13.9	5.6	5.5	3.7	4.9
Ethrel-1500 cc/ha	15.3	20.8	11.9	16.0	3.8	3.1	4.5	3.8
Ethrel-2000 cc/ha	13.7	18.6	10.9	14.4	4.9	4.0	4.9	4.6
Ethrel-2500 cc/ha	12.2	13.7	10.8	12.2	4.9	4.2	5.0	4.7
Control	25.0	26.9	13.2	21.7	3.5	3.0	2.0	2.8

\*DAS = Days after sowing.

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1) <sup>245</sup> New breeding lines of soybean developed at Pantnagar,

The major breeding objectives of the soybean breeding project at this university have been: high seed yield, early maturity, better seed quality and resistance particularly to yellow mosaic virus, bacterial pustule, and *Rhizoctonia* aerial blight. The suitable donors have been identified and are being used in the crossing program (Ram et al., 1981).

A total of 270 newly selected breeding lines from the advanced generations derived from the crossing program (pedigree method of breeding) were evaluated in 15 different trails, each consisting of 18 new breeding lines and 2 checks, i.e., 'Alankar' and 'Bragg'. The trials were planted on June 27, 1979, in randomized block designs with 2 replications. Each plot consisted of 5 rows of 5 m, spaced 60 cm. Detailed observations were recorded on incidence of diseases and agronomic characteristics. Based on overall superiority, plant type and freedom from diseases, 90 lines (PK-412 to PK-501) were selected for further evaluation in the next season.

These 90 new breeding lines were evaluated in 5 separate trials, each comprising 18 new lines and 2 checks (Bragg and Alankar). All these trials were planted on June 28, 1980, in randomized block design with 4 replications. The planting details were as given above.

The yield differences among the lines in trial 1 were significant. The lines giving better yield than the checks were PK-412, PK-413, PK-415, PK-416, PK-422, PK-424, PK-428 and PK-429. The highest yielding line in this trial was PK-422 (2916 kg/ha). The maturity duration of these lines ranged from 117-123 days.

The lines included in trial 2 did not have significant differences for seed yield. The highest yielding line in this trial was PK-430 (2794 kg/ha). Maturity duration ranged from 113 to 118 days.

The highest yielding line in trial 3 was PK-450 (3194 kg/ha). Other superior lines in this trial were PK-448, PK-449, PK-451, PK-454, PK-455, PK-459, PK-460, PK-463 and PK-464. The maturity duration of these lines ranged from 115 to 123 days.

The lines giving better yield than the checks in trial 4 were PK-467, PK-469, PK-470, PK-471, PK-472, PK-477 and PK-478. They had a maturity range of 119-125 days. Except for 3 lines (PK-484, PK-487, PK-501), these 15 lines gave more seed yield than the checks in trial 4. The maturity duration was 118-125 days.

The performance of these selected lines is given in Table 1. Most of these lines were resistant to yellow mosaic and bacterial pustule. The resistance to yellow mosaic has come from either UPSM-534 or *Glycine formosana*. Bragg was the source of resistance to bacterial pustules. Some of these lines and a few additional ones as given below have been included in the all-India coordinated testing:

Table 1. Performance of new breeding lines of soybean during rainy season 1980 at Pantnagar

Breeding lines	Parentage	Days to flowering	Days to maturity	Plant height (cm)	Pods/plant	Seeds/pod	100-seed weight (g)	Seed yield (kg/ha)	Disease reaction	
									YMV	Bp
1	2	3	4	5	6	7	8	9	10	11
PK-412	(M534 x S-38)	54	120	73.3	96.8	2.08	13.6	2500	M	R
PK-413	(M534 x S-38)	45	121	71.8	69.6	2.01	12.9	2326	R	S
PK-415	(M534 x S-38)	45	119	67.7	84.2	2.01	14.5	2361	M	R
PK-416	(M534 x S-38)	45	123	74.5	91.6	1.92	16.3	2465	M	R
PK-422	(M534 x S-38) Bragg	69	119	66.6	86.9	2.05	14.3	2916	M	M
PK-424	(M534 x S-38) Bragg	46	119	75.9	75.0	2.12	15.2	2412	M	R
PK-428	(M534 x S-38) Bragg	44	119	57.6	69.0	2.11	13.7	2152	M	R
PK-429	(M534 x S-38) Bragg	45	120	60.6	91.8	2.00	12.8	2761	R	M
PK-430	(M534 x S-38) Bragg	44	118	54.3	75.8	2.20	15.3	2794	R	R
PK-448	(M534 x M-91) Bragg	42	115	63.5	71.1	2.00	12.7	2639	M	R
PK-449	(M534 x M-91) Bragg	43	117	66.8	62.4	1.80	12.7	2465	M	M
PK-450	(M534 x M-91) Bragg	44	115	66.9	81.1	2.00	13.0	3194	M	M
PK-451	(M534 x M-91) Bragg	46	117	70.4	99.2	2.10	13.4	2517	R	R
PK-452	(M534 x M-91) Bragg	48	120	72.6	69.6	1.90	17.9	2690	M	R
PK-454	(M534 x M-91)	48	119	68.7	59.1	2.00	14.5	2378	R	R
PK-455	(M534 x M-91)	51	122	71.8	64.5	2.00	16.5	2326	M	R
PK-459	(M534 x M-168) Bragg	47	123	62.9	73.4	1.90	14.5	2708	R	R
PK-460	(M534 x M-168) Bragg	46	121	68.0	83.7	1.90	14.0	2430	R	R
PK-463	(Hardee x Pb-1)	55	118	83.2	86.9	2.00	13.4	2517	M	R
PK-464	(Hardee x Pb-1)	47	115	53.6	77.6	2.00	15.0	2378	M	R
PK-467	(Hardee x Pb-1)	58	120	72.7	80.5	2.05	12.80	2430	M	R
PK-469	(Hardee x Pb-1)	57	119	72.4	88.3	1.90	14.00	2430	M	R
PK-470	(Hardee x Pb-1)	57	119	72.4	74.0	2.00	13.6	2586	M	M
PK-471	(Hardee x Pb-1)	56	119	74.4	85.3	2.15	15.1	2812	M	R
PK-472	(Hardee x Pb-1)	61	125	62.5	90.1	1.95	15.7	3037	R	R
PK-477	(M534 x Pb-1)	57	124	99.3	94.2	1.95	12.2	2621	M	R
PK-478	(M534 x PK-71-39)	50	123	63.4	84.1	2.05	18.3	2430	R	M

Table 1. Continued

Breeding lines	Parentage	Days to flowering	Days to maturity	Plant height (cm)	Pods/ plant	Seeds/ pod	100-seed weight (g)	Seed yield (kg/ha)	Disease reaction
1	2	3	4	5	6	7	8	9	YMV 10 BP 11
PK-485	(M534 x PK-71-39) Bragg	47	119	123.2	97.9	1.90	15.7	2760	R R
PK-486	(GF x Bragg) Bragg	52	119	65.3	87.6	1.90	11.2	2100	R R
PK-488	(GF x Bragg) Bragg	52	120	70.3	76.0	2.05	13.0	2030	R R
PK-489	(GF x Bragg) Bragg	52	120	71.1	87.4	1.90	12.7	1961	R R
PK-490	(GF x Bragg) Bragg	52	119	66.2	91.6	1.95	13.7	2187	R R
PK-491	(GF x Bragg) Bragg	52	119	71.0	78.4	2.00	12.0	2014	R R
PK-492	(M534 x Lee)	52	117	55.3	74.1	2.15	17.4	2100	M M
PK-493	(T-49 x Lee)	52	123	73.7	79.2	1.80	15.5	2378	R R
PK-494	(T-49 x Lee)	51	119	65.9	88.2	2.15	11.2	2042	M R
PK-495	(M726 x T-49)	61	125	76.8	85.4	1.95	12.9	1892	M R
PK-496	(M726 x T-49)	61	123	93.8	86.7	1.85	16.3	2257	M R
PK-497	(M726 x T-49)	61	120	86.2	84.3	2.00	17.0	1926	M S
PK-498	(MS-2 x M534)	51	120	93.3	90.5	1.95	15.1	2621	M R
PK-499	(M534 x S-38) Bragg	50	120	69.1	67.5	2.00	16.1	2361	MR R
PK-500	(M534 x S-38)	57	120	58.0	87.9	1.85	20.1	2308	S R

M 534 = UPSM 534, YMV = Yellow mosaic virus, BP = Bacterial pustule, R = Resistant, M = Moderately resistant, S = Susceptible



Northern Hill Zone - PK-415, 429, 430, 442, 444, 450.  
 Northern Plain Zone - PK-412, 416, 448, 451, 453, 459, 478, 486, 490.  
 Central Zone - PK-395, 472, 484, 493, 500.  
 Southern Zone - PK-398, 408, 470, 471, 485, 498.

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Ram, H. H., Pushpendra, K. Singh and V. D. Verma. 1981. Breeding soybean varieties for the northern India. Soybean Genet. Newsl. 8:74-78.

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#### 2) <sup>245</sup> Extent of selfing during crossing in soybean,

Soybean is a strictly self-pollinated crop and crossing between two varieties is rather difficult due to small size of flowers and low pod setting (Ram et al., 1981). The crossed pods usually have reduced size and, hence, have fewer seeds/pod. The seeds obtained from the crossed pods may include some seeds that might be due to selfing while crossing. Our observations on the  $F_1$  generations in our breeding program clearly support this possibility. We invariably encounter selfed plants in the  $F_1$  generations. However, in this report, we intend to provide the extent of selfed seeds separately in single-seeded, double-seeded and triple-seeded crossed pods.

Seventy-five  $F_1$ s were grown on July 3, 1981. Each cross was divided into 3 groups, viz., single-seeded crossed pods, double-seeded crossed pods and triple-seeded crossed pods. The seeds were grown group-wise in single row, 2 m long, spaced 60 cm. Total  $F_1$  plants across the 75 crosses were counted groupwise and the selfed plants were identified based on flower color, plant type, growth habit, size of leaflet, pubescence color (purple flower > white flower, indeterminate growth habit > determinate growth habit, incomplete dominance between narrow and broad leaflet, tawny pubescence > grey pubescence). The results are summarized in the following table.

Table 1. Percentage of selfed seeds during crossing in soybean

Single-seeded crossed pods			Double-seeded crossed pods			Triple-seeded crossed pods		
Total plants	Selfed plants	Selfed plants (%)	Total plants	Selfed plants	Selfed plants (%)	Total plants	Selfed plants	Selfed plants (%)
163	16	9.8	365	73	20.0	96	43	44.8

The percentage of selfed plants was lowest (9.8%) in single-seeded pods and highest (44.8%) in triple-seeded pods. Taking these values into account, it is suggested that  $F_1$  plants should be carefully inspected and selfed plants rogued out. As far as possible, seeds from triple-seeded pods should be avoided to grow due to higher percentage of selfing in these crosses. Further crosses should be planned in such a way so that dominant phenotype comes from the male parent for successful roguing. It would be safer to grow  $F_2$  of each  $F_1$  plant separately in view of high level of selfing (9.8 - 44.8%) during crossing in soybean.

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### 3) <sup>245</sup> Screening for photoperiod insensitivity under natural conditions in soybean [7].

Early <sup>✓</sup>[varieties] of soybean have been found to be less sensitive to photoperiods than late varieties (Johnson et al., 1960). Therefore, it was postulated that some of the early strains of soybean may have no photoperiod requirement and accordingly screening for insensitivity to photoperiod was carried out in 498 early lines of soybean. These germplasm lines were evaluated for days to flowering and several morphological traits under two different seasons, viz., rainy season, 1978, and spring/summer season, 1979.

The difference in delay of days to flowering between rainy season and spring season plantings ranged from -4 to 40 days. These lines were classified into different groups (Table 1) according to the degree of delay in flowering during spring/summer season following the procedure of Shanmugasundaram (1978).

Table 1. Classification of lines into different groups based on delay in days to flowering under spring planting

Delay in days to flowering	Sensitivity score	Number of lines
-4-4	0	60
5-8	1	83
9-16	2	253
17-24	3	92
25-32	4	9
33-40	5	1
		<u>498</u>

The lines having a sensitivity score of 0 were considered as insensitive. These lines were as follows:

UPSE 6, 7, 75, 98, 104, 158, 164, 171, 175, 204, 339, 704, 2411,  
2619, 2628, 2631, 2632, 2673, 2678, 2687, 2690, 2718, 2723,  
2727, 2747, 2769, 2770, 2782, 2783, 2787, 2789, 2791, 2794,  
2795, 2798, 2799, 2800, 2802, 2803, 2806, 2808, 2813, 2819,  
2820, 2821, 2826, 2828, 2837, 2841, 2842, 2843, 2846, 2847,  
2848, 2849, 2893, 2896, 2897, 2900, 2913, 2914, 2937, 2948.

These lines may be grown throughout the country and may possibly be used as donors for photoperiod insensitivity. However, these lines when planted under Pantnagar conditions tend to flower too soon and, therefore, have low yields. Therefore, from yield point of view, these lines are inferior. Therefore, attempts are in progress to identify photoperiod insensitive, late maturing cultivars which may be utilized for breeding soybean in the tropics.

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245  
 15 Genotype x Environment interaction in soybean; I. Individual regression analysis,

In this communication, the results obtained from the material given earlier (Gupta et al., 1981) on the basis of individual regression analysis by using Perkins and Jinks (1968) model, have been presented for different traits groupwise. In the individual regression analysis, genotypes having nonsignificant regression m.s. as well as remainder m.s. against error m.s. were described as exhibiting absence of genotype x environment (g x e) interaction, genotypes having regression m.s. significantly different from error m.s. were designated as showing predictable g x e (linear g x e) interaction, and genotypes having only significant remainder m.s. or significant regression m.s. not significantly different from significant remainder m.s. were categorized as genotypes showing unpredictable g x e (nonlinear g x e) interaction.

On the basis of this all 40 genotypes were classified into three groups and their distribution with respect to different groups of characters studied is given in Table 1.

Seed yield and its components. In this group of characters, none of the genotypes showed absence of g x e interaction. Majority of the genotypes (about 60% genotypes for pods per plant, pods per main stem and pod length; 80% for seed yield per plant and 92.5% for seeds per pod) exhibited predictable g x e interaction.

Seed quality traits. For percent laboratory germination, percent field emergence and percent hard seed, 10, 18 and 23 genotypes showed absence of g x e interaction; 20, 6 and 16 genotypes exhibited predictable g x e interaction; while 8, 10, 16 and 7 genotypes had unpredictable g x e interaction, respectively. For 100-seed weight and 100-seed volume, none of the genotypes exhibited absence of g x e interaction, 55% genotypes showed predictable g x e interaction. For seed density and seed specific gravity index, majority of the genotypes exhibited unpredictable g x e interaction.

Structural components. None of the genotypes showed absence of g x e interaction for primary branches per plant and nodes per main stem, while only a few genotypes (about 10%) did so for rest of the traits. More than 50% of the genotypes exhibited unpredictable g x e interaction for plant height, primary branches per plant and nodes per main stem. Approximately 80% of the genotypes exhibited predictable g x e interaction for internode length and petiole length.

Phenological traits. None of the genotypes exhibited absence of g x e interaction for these traits. Almost all the genotypes (97.5%) indicated predictable g x e interaction for days to maturity while about half of them (52.5%) did so for days to first flowering.

Physiological traits. Approximately 80% of the genotypes had predictable g x e interaction and 10% exhibited absence of g x e interaction for pod potential per node, the only character studied in this group.

Table 1. Distribution of 40 genotypes on the basis of presence (or absence) and nature of genotype x environment interaction for different groups of characters

Character	G x E absent	Predictable G x E	Nonpredictable G x E
<u>Seed yield and its components</u>			
Seed yield/plant (gm)	0	32	8
Pods/plant	0	25	15
Pods/main stem	0	23	17
Pod length (cm)	0	26	14
Seeds/pod	0	37	3
<u>Seed quality traits</u>			
Percent lab. germination	10	20	10
Percent field emergence	18	6	16
Percent hard seed	23	10	7
100-seed weight (gm)	0	22	18
100-seed volume (cc)	0	22	18
Seed density (gm/cc)	4	2	34
Seed specific gravity index	2	11	27
<u>Structural components</u>			
Plant height (cm)	2	15	23
Branches/plant	0	17	23
First internode length (cm)	4	32	4
Petiole length (cm)	6	33	1
Nodes/main stem	0	19	21
<u>Phenological traits</u>			
Days to first flowering	0	21	19
Days to maturity	0	39	1
<u>Physiological traits</u>			
Pod potential/node	3	33	4

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2) <sup>245</sup> Genotype x Environment interaction in soybean; II. Joint regression analysis

The individual regression analysis has been given in the previous communication. In this article the results obtained from joint regression analysis (Table 1) according to Perkins and Jinks (1968) for all the groups of characters except leaf potential and leaf area where conventional  $g \times e$  interaction analysis was done, are being presented. The method and materials and the layout of the experiment was the same as reported by Gupta et al. (1981). The groupwise results obtained are given below.

Seed yield and its components. In this group, joint regression analysis indicated that all the items in the ANOVA (mean sum of squares for genotypes, environments, genotype x environments, heterogeneity among regressions and heterogeneity among deviations) except remainder m.s. were significant for seed yield per plant, pods per plant and pods per main stem, while for pod length and seeds per pod, m.s. due to genotypes as well as due to environments were significant. This indicated that genotypic differences existed for all the attributes studied in this group. Significant environmental m.s. for all the traits indicated the diversity present among the environments studied. The m.s. due to  $g \times e$  interaction was significant for all the traits except those for pod length and seeds per pod.

On partitioning the  $g \times e$  interaction into linear and nonlinear components, heterogeneity among regression (linear portion of  $g \times e$  interaction) was significant for seed yield, pods per plant and pods per main stem, while heterogeneity among deviations (nonlinear portion of  $g \times e$  interaction) was not significant, when tested against pooled error m.s., indicating, thereby, the presence of only linear (predictable)  $g \times e$  interaction.

Seed quality traits. Genotypic differences existed for all the traits studied in this group except percent field emergence. Diversity among environments was also present. All the traits except percent field emergence exhibited presence of  $g \times e$  interaction. Linear  $g \times e$  interaction was present for percent laboratory germination and percent hard seed, whereas for 100-seed volume, although linear  $g \times e$  interaction was predominant, yet nonlinear  $g \times e$  interaction was appreciable as indicated by significant heterogeneity among regression m.s. from significant remainder m.s. For 100-seed weight, seed density and seed specific gravity index, both linear and nonlinear  $g \times e$  interactions were equally important.

Structural components. Genotypic differences existed for all the components studied except internode length. Environments studied also differed significantly from each other. All the components except internode length exhibited the presence of  $g \times e$  interaction. Linear  $g \times e$  interaction was recorded for petiole length and nodes per main stem, whereas for primary branches per plant, though linear  $g \times e$  interaction was present, an equal amount of nonlinear  $g \times e$  interaction was also observed. However, for plant height, absence of linear  $g \times e$  interaction, and the presence of nonlinear  $g \times e$  interaction was observed as indicated by nonsignificant heterogeneity among regression m.s. and significant remainder m.s. from error m.s.

Phenological traits. For both the traits studied in this group, namely, days to first flowering and days to maturity, joint regression analysis indicated significant differences among genotypes and environments and presence of  $g \times e$  interaction. Linear as well as nonlinear  $g \times e$  interaction were equally important for both the traits.



Table 1. Joint regression/ genotype x environment/ interaction analysis in respect to different groups of characters

Source of variation	Mean sum of squares						
	1	2	3	4	5	6	7
<u>Seed yield and its components</u>							
Seed yield/plant (gm)		17.92*	2726.01*	9.50*	11.40*	8.87*	6.8855
Pods/plant		239.78*	13629.62*	86.36*	140.56	68.29	62.9762
Pods/main stem		28.07*	829.92*	6.58*	11.00*	5.11	4.5592
Pod length (cm)		0.22*	11.77*	0.087	0.11	0.08	0.0865
Seeds/pod <sup>+</sup>		0.076*	6.45*	0.20	0.026	0.016	0.277
<u>Seed quality traits</u>							
Percent lab germination		130.80*	3631.67*	90.17	122.75*	79.31	71.8709
Percent field emergence		260.27	1741.64*	175.20	154.94	181.96	193.2397
Percent hard seed		17.45*	202.34*	11.69*	30.32*	5.48	5.8728
100-seed weight (gm)		7.92*	181.04*	0.89*	1.05* <sup>@</sup>	0.84*	0.4305
100-seed volume (cc)		5.30*	132.20*	0.61*	0.81* <sup>@@</sup>	0.54*	0.2861
Seed density (gm/cc)		0.00071*	0.0031*	0.00025*	0.00027* <sup>@</sup>	0.00024*	0.00007
Seed specific gravity index		591.52*	7146.03*	100.53*	119.27* <sup>@</sup>	94.28*	58.7431
<u>Structural components</u>							
Plant height (cm)		179.30*	1322.77*	23.66*	19.27	25.12*	17.4215
Branches/plant		2.63*	116.58*	0.96*	1.14* <sup>@</sup>	0.90*	0.5183
First internode length (cm) <sup>+</sup>		0.50	50.32*	0.35	0.41	0.32	0.3809

Table 1. Continued

1	2	3	4	5	6	7
<u>Structural components (contd.)</u>						
Petiole length (cm) <sup>‡</sup>	20.69*	2523.96*	16.96*	24.85*	9.06	8.7039
Nodes/main stem	6.01*	230.66*	0.90*	1.04*	0.85	0.6865
<u>Phenological traits</u>						
Days to first flowering <sup>‡</sup>	24.93*	2381.87*	6.26*	7.15*@	66.18*	3.0619
Days to maturity	14.05*	3740.67*	3.29*	3.41*@	3.28*	1.4869
<u>Physiological traits</u>						
Pod potential/node <sup>+</sup>	0.20	36.16*	0.18	0.29*	0.12	0.1523
Leaves potential/node <sup>§</sup>	170.26*	30599.66*	54.77*	--	--	2.5215
Leaf area <sup>§</sup>	55.2248*	2470.8645*	57.0367*	--	--	3.3361

\*Significant at the 5% level.

@Heterogeneity between regressions significant against error M.S. but not against significant remainder M.S.

@@Heterogeneity between regressions significant against error M.S. as well as against significant remainder M.S.

<sup>+</sup>Data analyzed over 4 locations.

<sup>‡</sup>Data analyzed over 3 locations.

<sup>§</sup>Data analyzed over 2 locations.

## Degrees of freedom for joint regression/GxE interaction analyses

Source of variation	Characters			
	Analysis over 5 environments	Analysis over 4 environments	Analysis over 3 environments	Analysis over 2 environments
Genotypes	39	39	39	39
Environments (joint regression)	4	3	2	1
Genotypes x environments (G x E interaction)	156	117	78	39
Heterogeneity between regressions	39	39	39	---
Remainder	117	78	39	---
Error	195	156	117	78

Physiological traits. Among the three physiological traits studied, genotypic differences were significant for leaf area and leaf potential per plant but not significant for pod potential per node. However, g x e interaction was present for all the three traits. Linear g x e interaction was present for pod potential per node, whereas for leaf area and leaf potential per plant, nature of g x e interaction could not be studied as these were recorded only in two of the environments.

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3) <sup>245</sup> Association among productivity, responsiveness and stability for different groups of traits in soybean [12]

In <sup>2</sup> [India] soybean adaptability trials have revealed that, in order to stabilize yield and popularize soybean cultivation, breeders must look for <sup>1</sup> [genotypes] with good germinability and wider adaptability under diverse geographical and climatological situations (Singh, 1976). Prospects of developing different genotypes having varying degrees of adaptability would depend to some extent on the relationship between various adaptability parameters; namely,  $d_i$  (additive genetic effect-productivity),  $\beta_i$  (genotypic regression - a measure of responsiveness),  $S_d^{-2}$  (deviation from regression - a measure of stability). This has been attempted in the present study on the same material as reported earlier (Gupta et al., 1981) by working out the correlations over all the 40 genotypes between the three parameters which were calculated as per Perkins and Jinks (1968) taken in pairs (Table 1).

As per the results (Table 1) there appears to be a strong positive association between mean performance of a variety or its additive genetic effect ( $d_i$  in Perkins and Jinks, 1968 model) and ability to respond to a better environment which is particularly marked in case of seed yield per plant, pods per plant, pods per main stem, plant height, primary branches, petiole length, nodes per main stem, days to maturity and pod potential per node. Such a positive correlation for majority of yield components and structural components has also been documented earlier in different crops (Eberhart and Russell, 1966; Perkins and Jinks, 1968; Breese, 1969; Gupta et al., 1974; Langer et al., 1979). However, in soybean, on this association conflicting results have been reported. Verma et al. (1972) have reported positive association for majority of yield and structural components, but no association for days to maturity, whereas Smith et al. (1967), Tsai et al. (1967) and Walker and Fehr (1978) have reported apparently no association between mean yield and responsiveness. Moreover, this association for petiole length, nodes per main stem and pod potential per node has been documented only for the first time in the present study.

In respect to all the seed quality traits, namely 100-seed weight, 100-seed volume and seed density, though apparently there is a positive correlation between ' $d_i$ ' and ' $\beta_i$ ' but it is not significant. For seed specific-gravity index, absence of such a relationship has been reported in soybean by Verma et al. (1972) and for some of the quality characters in chickpea by Gupta et al. (1974).

Among seed yield and its components, there is no apparent relationship between ' $d_i$ ' and ' $\beta_i$ ' for pod length and seeds per pod and same is the case for days to first flowering. In literature, a significant positive association has been reported for days to first flowering, 100-seed weight, and pod length, but no association for days to maturity, which is in contradiction with the present findings but for seeds per pod and seed specific-gravity index, the present results are in conformation with earlier reports (Verma et al., 1972).

There is no apparent association of stability parameter  $S_d^{-2}$  with mean performance ( $d_i$ ) and responsiveness ( $\beta_i$ ) for majority of the characters studied including seed yield, though the trend is weak and becomes positively significant only in case of pods per plant, pods per main stem, pod length and primary branches per plant with respect to ' $d_i$ ' and ' $S_d^{-2}$ ', and it becomes

positively significant for nodes per main stem and negatively significant for pod length, 100-seed weight and primary branches per plant with respect to  $\beta_i$  and ' $S_d^{-2}$ '. The information on the association of ' $S_d^{-2}$ ' with  $d_i$  and  $\beta_i$ , in the literature for soybean is not available. However, more or less similar association, as recorded in present study, has been reported in chickpea by Gupta et al. (1974) and in oats by Langer et al. (1979). Hence, these associations reveal that, for major yield components, structural components and maturity, a breeder can simultaneously select both for high yield and high responsiveness to changing environments. For seed quality traits, also, situation is equally good, because one can have specific combination of desired nature, like high seed quality and least responsive or high seed quality and average responsive or high seed quality and above-average responsiveness. Independent behavior of ' $S_d^{-2}$ ' in relation to other adaptability parameters provides a happier situation for combining high stability with high productivity ( $d_i$ ) and high productive response ( $\beta_i$ ) for majority of economic traits, e.g., seed yield, seed density and seed specific-gravity index. However, the positive association of ' $d_i$ ' and ' $S_d^{-2}$ ' with respect to pods per plant, pods per main stem, pod length and primary branches per plant has to be viewed cautiously. The negative association between ' $\beta_i$ ' and ' $S_d^{-2}$ ' for 100-seed volume is useful from a breeding point of view.

Table 1. Estimates of correlation coefficients between various adaptability parameters ( $d_i$ ,  $\beta_i$  and  $S_d^{-2}$ ) for different groups of characters

Characters	Correlation between		
	$d_i$ and $\beta_i$	$d_i$ and $S_d^{-2}$	$S_d^{-2}$ and $\beta_i$
<u>Seed yield and its components</u>			
Seed yield/plant (g)	0.82*	0.04	-0.12
Pods/plant	0.86*	0.37*	0.29
Pods/main stem	0.88*	0.42*	0.21
Pod length (cm)	-0.07	0.38*	-0.32*
Seeds/pod	0.10	0.14	0.10
<u>Seed quality traits</u>			
100-seed weight	0.020	0.01	-0.26
100-seed volume	0.20	-0.09	-0.37*
Seed density	0.13	0.10	-0.06
Seed specific gravity index	0.20	0.02	0.03
<u>Structural components</u>			
Plant height (cm)	0.54*	0.18	-0.16
Primary branches/plant	0.71*	0.41*	0.35*
Petiole length (cm)	0.87*	0.09	0.18
Nodes/main stem	0.71*	0.21	0.39*
<u>Phenological traits</u>			
Days to first flowering	0.21	0.19	-0.08
Days to maturity	0.77*	0.11	0.06
<u>Physiological traits</u>			
Pod potential/node	0.48*	0.26	0.06

\*Significant at 5% level.



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## 4) Genetic control of productivity, responsiveness and stability for different groups of traits in soybean,

In order to breed adaptable varieties, selection is to be based on the above three parameters of adaptability simultaneously, for achieving the desired objectives. After understanding the association between these, it is imperative to have information on the gene action or genetic architecture of these three parameters, because in a self-pollinated crop like soybean where end product is homozygous and homogeneous population, selection will be fruitful only if gene action is of additive or additive x additive nature.



In the present study, an attempt has been made to infer the genetic control of mean performance (di-productivity), responsiveness and stability with respect to each different group of characters studied, on the basis of segregation pattern observed among 36  $F_4$ -derived lines, of the cross Soybean Pb.1 x D 60-9647, in relation to their parents. The other details of the experiment were the same as reported earlier (Gupta et al., 1981), except that the parameters of productivity (di), responsiveness ( $\beta_i$ ) and stability ( $S_d^{-2}$ ) were estimated as per Perkins and Jinks (1968a,b). It can be seen from Table 1 that in general for seed yield and its components, performance appears to be under additive type of gene action as the majority of the segregants are having parent-dependent mean performance. The occurrence of transgressive segregants, having mean performance more than the parents for most of the yield components might be due to higher order of additive x additive epistatic interaction. Such a genetic control of mean performance has also been earlier documented in soybean (Leffel and Weiss, 1958; Leffel and Hanson, 1961; Paschal and Wilcox, 1975; Bhatade et al., 1977).

The segregants for this group of traits (seed yield and its components) have also shown parent-dependent responsiveness, as indicated by very high frequency of segregants having average responsiveness when the two parents are also average responsive. For this parameter ( $\beta_i$ ) also, seed yield and its components have thrown transgressive segregants though frequency is low as compared to mean performance. From this, it can be inferred that genetic control for responsiveness is also of additive x additive nature.

It is interesting to note that, for seed yield and its components, the segregants exhibit parent-dependent stability ( $S_d^{-2}$ ). For seed yield per plant and seeds per pod, both the parents were having high stability and more than 75% of the segregants have also exhibited stability. For pods per main stem, both the parents were unstable and it is interesting to see that for this character, approximately 50% of segregants are stable and 50% are unstable. For pods per plant and pod length, where only one of the parents is stable, approximately 60% of the segregants exhibit stability. For this parameter ( $S_d^{-2}$ ) though genetic control appears to be predominantly additive for seed yield and seeds per pod but for pods per plant, pods per main stem and pod length, genetic control might be due to additive, dominance and epistatic interactions as well. Therefore, it appears that at least selection is likely to be effective for all the three parameters of adaptability for seed yield and seeds per pod, a character of fitness.

For seed quality traits, parent-dependent mean performance and responsiveness observed for segregants indicate the predominant role of additive gene action. Transgressive segregants are apparent particularly for seed specific-gravity index. However, responsiveness for seed specific-gravity index appears to be under predominantly dominant type of gene action, as more than 90% of the segregants have shown average responsiveness as possessed by one of the parents. For seed quality traits, also, stability ( $S_d^{-2}$ ) appears to be under complex genetic control. However, for percent laboratory germination and percent hard seed, stability appears to be due to dominance as 90% of the segregants have stable behavior as shown by one of the parents. Therefore, for percent laboratory germination, selection may be effective for the three parameters, but for other seed quality traits, though selection might be profitable for mean performance and responsiveness but may not be so for high stability.

For structural, phenological and physiological traits also, performance and responsiveness appear to be parent-dependent, indicating thereby additive type of genetic control but stability parameter seems to be under complex genetic control. In literature, at least for soybean, the information on genetic control of responsiveness and stability parameter is limited except Oka (1973), where he has depicted yield stability as genotypic specific in some of Taiwanese varieties. However, some reports are available on the genetic control of mean performance with respect to some yield and developmental components but for germinability relevant information is not available (Leffel and Weiss, 1958; Weber et al., 1970; Paschal and Wilcox, 1975; Srivastava et al., 1978).

Some studies reported on other crops show that responsiveness and stability, at least to some extent, are under genetic control (Perkins and Jinks, 1968, a & b; Bucio-Alanis et al., 1969; Paroda and Hays, 1971) and the mean performance and linear sensitivity can be predicted successfully from one generation to another of the same cross, and it has been actually observed in the present study, too, with respect to various characters. Gupta (1971) in chickpea and Bains (1976) in wheat have also shown parent-dependent mean performance, responsiveness and stability in segregating generations.

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Table 1. Pattern of segregation with respect to productivity (di), responsiveness ( $\beta$ i) and stability ( $S^2_d$ ) of 36  $F_4$ -derived lines in relation to their parents

Characters	Productivity				Responsiveness				Stability					
	Parental		Distribution of segregants		Parental		Distribution of segregants		Parental		Distribution of segregants			
	P <sub>1</sub>	P <sub>2</sub>	BA	A	AA	P <sub>1</sub>	P <sub>2</sub>	BA	A	AA	P <sub>1</sub>	P <sub>2</sub>	Stable	Unstable
	P <sub>1</sub>	P <sub>2</sub>	BA	A	AA	P <sub>1</sub>	P <sub>2</sub>	BA	A	AA	P <sub>1</sub>	P <sub>2</sub>	P <sub>1</sub>	(+)
1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Seed yield and its components														
Seed yield/plant (g)	A	A	1	32	3	A	A	0	34	2	.	.	26	10
Pods/plant	A	A	4	29	3	A	A	2	32	2	.	+	23	13
Pods/main stem	BA	A	5	24	7	A	A	1	31	4	+	+	17	19
Pod length (cm)	A	A	3	29	5	A	A	2	34	0	.	+	22	14
Seeds/pod	A	A	5	21	10	A	BA	1	32	3	.	.	34	2
Seed quality traits														
Percent laboratory germination	A	A	2	33	1	A	A	1	33	2	.	+	27	9
Percent hard seed	A	A	0	31	5	A	A	6	24	6	+	.	29	7
100-seed weight (g)	BA	AA	6	21	9	A	A	0	35	1	+	+	12	24
100-seed volume (cc)	BA	AA	5	22	9	A	A	0	35	1	+	+	14	22
Seed density (g/cc)	A	AA	5	29	2	A	A	2	33	1	+	+	7	29
Seed specific gravity index	A	A	8	22	6	BA	A	2	33	1	.	.	11	25
Structural components														
Plant height (cm)	A	A	7	24	5	A	A	1	34	1	+	.	15	21
Primary branches/plant	A	A	3	27	6	A	A	2	31	3	+	.	12	24
Petiole length (cm)	A	A	1	33	2	A	A	5	27	4	.	.	34	2
Nodes/main stem	A	A	7	20	9	A	A	1	33	2	+	+	16	20
Phenological traits														
Days to first flowering	AA	BA	11	15	12	A	A	1	34	1	.	.	19	17
Days to maturity	A	A	5	27	4	A	A	1	34	1	+	+	14	22



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5) Factor analysis in  $F_2$  generation of soybean crosses.

Factor analysis is a technique of reducing a large number of correlated variables to a few main factors. It has been resorted to to overcome the limitations of univariate methods of analysis like correlations, path-coefficient and regression analysis (Wright, 1960; Walton, 1972). Besides, Moreno and Cubero (1978) have used it for estimating diversity. This paper purports to report findings of factor analysis in soybean crosses to know important traits for yield selection and diversity among crosses.

Materials and methods: The  $F_2$  populations of soybean crosses 'Hill' x 8-3, 'Davis' x 8-3, 'Lee' x 8-3, and 'Semmes' x 8-3, along with the parents, were grown in a randomized block design with four replications at the Indian Agricultural Research Institute, New Delhi. The varieties used as female parents are medium-tall, except Semmes (tall), and adapted in the north Indian plains and hills, whereas the common male parent, a tall variety, gives better performance in the southern parts of the country. Each plot (5m x 3m) had row-to-row and plant-to-plant spacing of 45 and 5 cm, respectively. Data on plant height, branches/plant, pod clusters/plant, pods/plant and seed yield/plant were recorded from 50 randomly selected plants from each replication. The data were tested for the existence of variability and significant traits were correlated in all possible combinations at phenotypic level. These correlations were used for factor analysis through principal component method, as suggested by Harman (1968). The analysis was terminated after the factors accounting for more than 90% variability were extracted.

Results and discussion: Phenotypic correlations (Table 1) indicated that correlations among various traits resembled across crosses except plant height in Semmes x 8-3 cross, which were not significant. The results showed that in medium-tall x tall populations improvement of seed yield and yield components is likely to prove effective with the selection of taller plants. Branches, pod clusters and pods/plant showed significant correlation with seed yield/plant in all the crosses.



Table 1. Phenotypic correlations in the F<sub>2</sub> generation of soybean crosses

Character	Cross	Branches/ plant	Pod clusters/ plant	Pods/ plant	Seed yield/ plant (g)
Plant height (cm)	Hill x 8-3	0.373*	0.583*	0.422*	0.343*
	Lee x 8-3	0.282*	0.482*	0.372*	0.294*
	Davis x 8-3	0.328*	0.405*	0.256*	0.287*
	Semmes x 8-3	0.125	0.107	0.129	0.136
Branches/plant	Hill x 8-3		0.795*	0.737*	0.640*
	Lee x 8-3		0.782*	0.719*	0.650*
	Davis x 8-3		0.721*	0.662*	0.446*
	Semmes x 8-3		0.757*	0.676*	0.604*
Pod clusters/plant	Hill x 8-3			0.866*	0.722*
	Lee x 8-3			0.868*	0.745*
	Davis x 8-3			0.901*	0.692*
	Semmes x 8-3			0.886*	0.833*
Pods/plant	Hill x 8-3				0.830*
	Lee x 8-3				0.838*
	Davis x 8-3				0.694*
	Semmes x 8-3				0.827*

\*Significant at the 1% level.



Table 2. Factor loadings in soybean crosses

Character	Crosses							
	Hill x 8-3		Lee x 8-3		Davis x 8-3		Semmes x 8-3	
	Factors		Factors		Factors		Factors	
	I	II	I	II	I	II	I	II
Plant height	0.599	-0.344	0.516	-0.331	0.478	-0.252	0.153	-0.109
Branches/plant	0.810	0.281	0.818	0.290	0.491	0.401	0.822	-0.219
Pod clusters/plant	0.949	0.063	0.808	0.137	0.999	0.355	0.936	0.153
Pods/plant	0.881	0.271	0.902	0.309	0.933	0.401	0.915	0.258
Eigen value	2.724	0.275	2.555	0.307	2.340	0.511	2.412	0.150
Explained variation (%)	68.111	6.875	63.884	7.697	58.500	12.778	60.312	3.745

Factor analysis (Table 2) indicated that plant height, branches/plant, pod clusters/plant and pods/plant recorded their highest factor loadings in all the crosses in Factor I. The only exception was Semmes x 8-3 (tall x tall), where height was not important for the improvement of the yield. It appears that, in this cross, height is being affected by some other factor. The first factor recorded the highest factor loading for all the traits under study in all the crosses, indicated their overwhelming importance over other traits. Though no weightage was given to yield in this analysis, all the crosses indicated that all yield components were affected by the most important factor, i.e., Factor I. The present study indicated that minimum of 0.282\*\* correlation among the traits was affected by a single factor. In case of Factor I, higher loadings were in favor of direct yield components (pods/plant and pod clusters/plant) and the lower loadings were in favor of growth traits (plant height and branches/plant), indicating that this factor was essentially related to the yield potential of plants. A comparison of the factor loadings in different crosses revealed variation in the size of the loadings although the composition of the variables in this factor remained essentially the same in all the crosses.

After the extraction of the first factor, the second factor did not appear to affect any trait except moderate effect on Davis x 8-3. In general, two factors accounted for 64-75% of the communality (proportion of total variance for phenotypic correlation matrix) in all the crosses. The results indicated that, even with a smaller number of the variables included in the study, factor analysis was potent in grouping four correlated variables into two factors. For the plant breeder, such information may well assist both by increasing his understanding of the relative importance of the yield components and growth traits and by helping to determine the nature and sequence of the traits for which the selections were to be made in his breeding program.

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6) Effect of environment and cropping system on the coefficients of variability for seed yield, quality, structural, phenological and physiological traits in soybean [ ]

Introduction. In the present investigation, the results obtained on the effects of environment and cropping system on the parameters of genetic variability for seed yield, quality, structural, phenological and physiological traits in soybean have been presented.

Materials and methods. The material, experimental design and characters studied were the same as reported by Gupta et al. (1981a). The data were further analyzed as per Johnson et al. (1955) for estimating various parameters of variability in different environments and cropping systems.

Results and discussion. Estimates of coefficient of variation at genotypic and phenotypic levels for different groups of characters over environments and cropping systems are given in Table 1.

Effect of cropping system. For seed yield per plant, genotypic, as well as phenotypic coefficients of variation were more in Palampur intercropping than in Palampur monoculture. For pods per plant, coefficients of variation (PCV and GCV both) were almost at par in the two cropping systems. For other three yield components, namely, pods per main stem, pod length and seeds per pod, GCVs were higher in case of Palampur monoculture than in Palampur intercropping, but converse was true for PCVs.

Among seed quality traits, for germination indices, percent laboratory germination, percent field emergence and percent hard seed, both PCV as well as GCV were higher in intercropping except percent hard seed where PCV was more in monoculture. Both for 100-seed weight and 100-seed volume, GCV and PCV were reduced in intercropping system, reduction being more in GCV. For seed density, GCV was more in monoculture but PCV was high in intercropping. For seed specific gravity index, estimates of GCV and PCV were larger in monoculture than in intercropping system.

Among structural components, GCV was substantially reduced in intercropping for primary branches per plant, first internode length and nodes per main stem. Both GCV and PCV were reduced for plant height in intercropping, though magnitude of reduction was low. PCV was also low in intercropping for other three traits except for primary branches per plant, where it was more. For days to maturity, GCV and PCV, both, were higher in monoculture than in intercropping. Drastic reduction was observed for the estimate of GCV for pod potential per node in intercropping but PCV was higher.

Effect of environment. As reported earlier (Gupta et al., 1981a), the environments sampled were varying with respect to altitude, and other climatic factors like rainfall and temperature. It can be seen from Table 1 that, in lower altitude (Kangra), GCV was considerably increased, while at Katrain, which is at a higher elevation, considerable reduction in the GCV was observed for seed yield. However, the PCV remained more or less stable over environments. For other seed yield components, namely pods/plant, pod length and seeds/pod, though PCV remained unchanged irrespective of altitude except with slight upward value in lower altitude, the GCV fluctuated considerably over environments. However, among seed yield components, pods per main stem was the least influenced by environments with respect to GCV.

In case of seed quality attributes, the effect of environment on both GCV and PCV was more pronounced in case of percent hard seed, specific gravity index, percent germination, whereas the estimates of other traits were least influenced. Among seed yield and seed quality components, a high magnitude of GCV was observed for percent hard seeds, specific gravity index and pods per main stem, in order.

In case of structural components, the magnitude of both GCV and PCV remained more or less stable over environments with the exception of primary branches per plant and petiole length. The PCV was enhanced at higher altitudes for primary branches, whereas it was reduced for petiole length.

In phenological traits, days to first flower exhibited stability for GCV over environments. At higher altitude, however, the estimates of these coefficients were comparatively reduced for days to maturity. For pod potential per node, the GCV estimates were drastically reduced at higher altitude, whereas PCV estimates were not influenced much. Among structural, phenological and physiological traits, petiole length, plant height, primary branches, exhibited comparatively high GCV estimates.

Conclusion. In general, both altitude and cropping system were found to influence the coefficients of variability both at phenotypic and genotypic level for different groups of traits in soybean. In the absence of limited reports available, on the influence of environments on the coefficient of variability, the above information would be valuable in breeding programs.

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Table 1. Estimates of coefficients of variation (c.v.) at genotypic and phenotypic levels for different groups of characters over environments

Characters	Environments					
		Palampur (mono- culture)	Palampur (inter- cropping)	Solan	Katraia	Kangra
<u>Seed yield and its components</u>						
Seed yield/ plant (gm)	G	15.66	21.75	18.20	4.99	27.41
	P	31.84	49.27	29.60	31.80	35.35
Pods/plant	G	9.99	10.93	25.76	18.57	7.52
	P	33.75	32.73	33.08	29.31	46.47
Pods/main stem	G	21.86	0.61	25.88	25.68	32.12
	P	31.72	37.41	35.53	29.57	58.49
Pod length (cm)	G	5.72	0.93	6.04	4.99	4.90
	P	10.36	21.51	8.79	8.51	7.56
Seeds/pod	G	4.54	2.04	4.09	--	5.52
	P	10.18	19.73	10.35	--	9.74
<u>Seed quality traits</u>						
Percent lab germination	G	1.69	5.54	3.49	9.62	6.87
	P	6.98	25.90	12.35	18.72	11.81
Percent field emergence	G	10.12	18.99	0.08	0.06	14.34
	P	49.29	67.29	48.56	44.80	49.35
Percent hard seed	G	70.50	138.75	149.79	81.99	82.72
	P	275.35	215.07	291.99	176.13	130.93
100-seed wt. (gm)	G	9.54	6.11	7.79	9.73	8.12
	P	10.79	9.41	8.64	11.03	10.69
100-seed volume (cc)	G	9.21	6.41	7.78	9.90	7.77
	P	10.56	9.55	8.65	11.04	10.19
Seed density (gm/cc)	G	1.33	1.29	1.15	1.59	1.44
	P	1.61	1.74	1.60	1.76	1.71

Table 1. Continued

Characters		Environments				
		Palampur (mono- culture)	Palampur (inter- cropping)	Solan	Katraian	Kangra
Seed specific gravity index	G	53.97	20.65	38.09	39.26	30.72
	P	69.34	37.75	56.65	46.52	37.08
<u>Structural components</u>						
Plant height (cm)	G	10.06	8.51	15.90	15.57	9.44
	P	16.91	16.53	19.19	17.79	16.45
Primary branches/ plant	G	11.34	1.23	12.49	12.34	22.57
	P	19.52	33.53	20.53	21.00	29.91
First inter- node length (cm)	G	4.65	0.84	0.49	--	5.30
	P	19.63	16.26	18.42	--	16.18
Petiole length (cm)	G	--	0.23	17.93	7.25	--
	P	--	19.55	28.16	13.39	--
Nodes/main stem	G	6.59	2.05	11.52	9.15	10.48
	P	13.16	12.10	15.56	11.31	12.92
<u>Phenological traits</u>						
Days to first flowering	G	7.18	--	7.85	--	6.49
	P	7.41	--	8.91	--	6.61
Days to maturity	G	1.93	1.50	1.58	0.51	2.64
	P	2.05	1.89	3.17	1.65	2.74
<u>Physiological traits</u>						
Pod potential/ node	G	10.23	1.52	0.81	--	17.72
	P	18.52	25.30	21.86	--	25.77

G denotes c.v. at genotypic level.

P denotes c.v. at phenotypic level.

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7) Consistency of heritability estimates over environments and cropping systems for different groups of traits in soybean

Introduction. Though numerous reports are available on heritability estimates from individual environment, yet information on consistency of heritability over environments is meager (Byth et al., 1969). An attempt has been made in the present communication to understand the influence of environments and cropping system on the heritability estimates in soybean.

Materials and methods. The materials and experimental design were same as reported by Gupta et al. (1981). Heritability estimates (broad sense) were calculated as ratio of genotypic variance to phenotypic variance, for different groups of traits over environments.

Results and discussion. Heritability estimates (broad sense) obtained from different environments and under intercropping system where soybean genotypes were raised in association with maize, are given in Table 1. As indicated above, there are very few reports on soybean on the influence of environment on heritability. Byth et al. (1969) evaluating genetically homogeneous lines obtained from two crosses in soybean, for nine characters in three environments, observed that heritability was relatively consistent for all traits except seed yield where heritability was enhanced under favorable growth conditions and reduced when moisture stress was alleviated. As the experimental material in the present study consisted of 40 entries representing  $F_4$  derived lines of an intervarietal cross, and four standard pure line varieties, it will be interesting to examine the results obtained on heritability estimates incorporated in Table 1. Among all the traits studied, days to first flowering showed consistently highest heritability across environments.

Heritability for seed yield was high at lower elevation (Kangra) and at higher elevation (Solan, Palampur and Katrain) it was so reduced that at Katrain, the genetic variation was not significant. Under intercropping system at Palampur, the heritability was reduced as compared to monoculture system. The present results are contrary to those reported by Byth et al. (1969), because, while evaluating the same material, Gupta et al. (1981) reported Solan and Katrain as the favorable environments and Palampur monoculture as well as intercropping and Kangra as unfavorable. Therefore, the present results indicate that there is no relationship between heritability estimates and environmental status and there was a considerable inconsistency in heritability estimates for yield over environments. More or less similar situation was observed with respect to consistency of heritability and environmental status for pods per plant, but in this case heritability was low at the lower elevation (Kangra). For pods/main stem, pod length and seeds/pod, though, there was not much influence of location on consistency of heritability estimates, yet there was drastic influence of intercropping system on heritability estimates as can be seen from Table 1. For these traits also, there was no apparent relationship of heritability estimates with environmental status.

Among seed quality attributes, the influence of altitude on heritability was more pronounced for percent laboratory germination and percent hard seed,

Table 1. Heritability estimates for different groups of characters over environments and cropping system in soybean

Character	Environments				
	Palampur monoculture	Palampur inter- cropping	Solan	Katraia	Kangra
<u>Seed yield and its components</u>					
Seed yield/plant (g)	24.2	19.5	37.8	2.5*	60.0
Pods/plant	8.8*	11.2*	60.6	40.2	2.6*
Pods/main stem	47.5	0.0*	53.1	75.4	30.2
Pod length (cm)	30.5	0.2*	47.3	34.4	42.0
Seeds/pod	19.9	1.1*	15.6	0.0*	32.1
<u>Seed quality traits</u>					
Percent lab. germination	5.9*	4.6*	7.9*	26.4	33.8
Percent hard seed	6.6*	41.6	26.3	21.7	39.9
100-seed weight (g)	78.1	42.1	81.2	77.4	57.7
100-seed volume (cc)	76.2	45.0	80.8	80.4	58.1
Seed density (gm/cc)	68.2	55.3	51.9	80.9	70.9
Seed specific gravity index	60.6	29.9	45.2	71.2	68.6
<u>Structural components</u>					
Plant height (cm)	35.4	26.5	68.6	76.6	32.9
Branches/plant	33.7	0.1*	37.0	34.5	56.9
Nodes/main stem	25.0*	2.9*	54.8	65.5	65.7
Petiole length (cm)	--	0.0*	40.5	29.3	--
<u>Phenological traits</u>					
Days to first flower	93.7	--	77.8	--	96.2
Days to maturity	89.2	62.7	24.7	9.43*	62.7
<u>Physiological trait</u>					
Pod potential/node	30.5	0.4*	0.1*	--	47.3

\*Nonsignificant differences among genotypes as per the analysis of variance in the respective environment.

whereas the drastic influence of cropping system on heritability was evident for all the quality traits, namely, percent lab. germination, percent hard seeds, 100-seed weight, 100-seed volume, seed density, and seed specific-gravity index. The environmental status had, however, no relationship with the heritability of quality traits as well. It is interesting to note that at the same location, the heritability of percent hard seeds has enhanced from

6.6% in monoculture to 41.6% in intercropping system and for other quality traits, the heritability was considerably reduced under intercropping system as compared to monoculture. In general, the heritability estimates with respect to 100-seed weight, 100-seed volume, seed density and specific-gravity index, were consistently high over locations.

For structural components, namely plant height, branches/plant, nodes/main stem and petiole length, though both locations and cropping system had influenced the heritability estimates, however, influence of the latter was much more, reducing the heritability to zero level. For these traits also there was no apparent relationship of the environmental status with heritability except for petiole length where favorable environment exhibited higher heritability.

For days to first flower, though, there was consistency in heritability over locations but the heritability estimates for days to maturity greatly fluctuated over environments and cropping system. For pod potential per node, both environments and cropping system influenced the heritability estimates.

Conclusion. In general, both environment and cropping system exhibited considerable influence for majority of the traits studied and there was no apparent relationship between heritability estimates and environmental status. With exception of percent hard seed, heritability estimates were considerably reduced under intercropping system as compared with monoculture.

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- 8) Association of leaf and root characteristics among themselves and with seed yield, structural, physiological and phenological traits in soybean [1]

Introduction. The variation present for leaf and root characteristics has been reported earlier and in this communication the association of these traits with seed yield and other traits is being presented.

Materials and methods. Materials used in this study and design of experiment were the same as reported by Gupta et al. (1981) and in the earlier communication on variation for leaf and root characteristics. The data were further analyzed for estimating the correlation coefficients at phenotypic and genotypic level.

Results and discussion. Though the correlation coefficients at genotypic, phenotypic and environmental level were estimated among root and leaf characteristics themselves and also with other traits in the material mentioned earlier (Gupta et al., 1981), the only significant correlation coefficients at phenotypic level have been given in Table 1, as the genotypic correlation coefficients, in general were higher than phenotypic correlations, indicating inherent association among various traits. It can be seen from Table 1 that, irrespective of the environment, leaf potential per plant is positively associated with pods per plant, primary branches per plant and nodes per main stem. Leaf potential per plant (a measure of source) has also shown a positive association with pod potential per node (a measure of sink) at Solan. However, pod potential per node could not be studied at Katrain. There is also positive association between leaf potential per plant and unit leaf area. Leaf area, interestingly, is also positively associated with yield, petiole length and plant height at both locations. However, both leaf potential per plant and leaf area have shown strong positive association with seed yield, pods per plant, plant height, petiole length, days to first flowering, days to maturity and pod potential per node at Solan. The association observed at Katrain has to be viewed cautiously because there is absence of genotypic differences for yield, leaf potential per plant and leaf area. Hence, if one considers the relationship of these characters at Solan, it can be concluded that there is a good correlation between source and sink in the present material and an ideal plant as described under ideotype for wider adaptability (Garg, 1979) also needs to have high leaf potential per plant and more unit leaf area, besides larger petiole responding to changing environments and higher number of nodes per plant, thus leading to high yield. Earlier reports also indicate varietal differences in net photosynthesis as a result of differences in total leaf area per plant (Ozima, 1972).

The larger petiole length and higher number of nodes would provide more aerial display for leaves and thus reduce the mutual shading effect due to high leaf potential and per-unit leaf area. The reduction in yield in spite of high leaf potential and leaf area has been recorded earlier mainly due to mutual shading of leaves (Wallace and Munger, 1965; Adam, 1975). The poor yield obtained for pine-shaped mutant having more petiole length at the base as compared to tips (Tattersfield and Williams, 1979) might be due to less leaf potential or less unit leaf area.

For the root characteristics studied in the present investigation, namely nodules per plant, nodule dry matter per plant and root dry matter per plant, the [varietal differences] appear to be marked due to larger environmental influences. However, all the three root characteristics are positively associated with seed yield, pods per plant, nodes per main stem, petiole length and days to first flowering.



Table 1. Significant associations of leaf and root characteristics with yield and other traits in soybean

Characters	Leaf characteristics (at Solan)		Root characteristics (at Palampur)-monoculture		
	Leaf potential/ plant	Leaf area	Nodules/ plant	Nodule dry matter/ plant	Root dry matter/ plant
Seed yield/plant (g)	0.65	0.40	0.31	--	0.37
Pods/plant	0.78	0.45	0.35	0.35	0.44
Pods/main stem	0.48	-- <sup>a</sup>	--	--	--
Plant height (cm)	0.56	0.43	--	--	--
Primary branches/plant	0.61	--	--	--	0.41
Petiole length (cm)	0.51	0.39	--	--	--
Nodes/main stem	0.54	--	0.41	0.40	0.46
Days to first flowering	0.39	0.39	0.38	0.35	0.49
Days to maturity	0.33	0.34	--	--	--
Pod potential/node	0.41	0.60	--	--	--
Leaf potential/plant	--	0.36	--	--	--
Leaf area	0.36	--	--	--	--
Nodules/plant	--	0.42	--	0.73	0.66
Nodule dry matter/plant	--	--	0.73	--	0.70
Root dry matter/plant	--	--	0.66	0.70	--

<sup>a</sup>Correlation not calculated or nonsignificant at 5%.

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95 245  
Variation and heritability for leaf and root characteristics in soybean, across locations;

Introduction. Generally yield of soybean has been reckoned in terms of yield components like pods per plant but it is only one aspect of an overall allometry for enumerating the soybean plant. The other components which need attention are the physiological and root characteristics, which remained ignored so far, in breeding programs (Adam, 1975). To surpass the yield plateau in soybean, it is imperative that we pay due attention to these traits with a view to increase the photosynthetic efficiency and translocation activities through the genetic improvement of leaf and root characteristics. Hence, the present study was conducted to obtain information on nature of variation and heritability of these traits in soybean.

Materials and methods. Materials used in this study and design of experiment were reported by Gupta et al. (1981), except that in this study, data were obtained only from two locations -- Solan and Katrain (for leaf potential per plant and leaf area), and from Palampur and Solan (for nodules/plant). Nodule dry weight and root dry weight were recorded only at Palampur. Leaf characteristics were recorded at pod-formation stage and root characteristics were taken at 50% flowering stage.

Results and discussion. Estimates of means, range, variances and coefficient of variation (phenotypic, genotypic, environmental) and heritability for leaf potential/plant leaf area, nodules per plant, nodule dry weight per plant and root dry weight per plant are given for locations in Table 1. The available information on leaf characteristics with respect to variation and heritability is limited, however, Lal and Haque (1972) reported high estimates of genotypic coefficients of variation for number of leaves per plant and total leaf area. The estimates of range, genotypic coefficient of variation and heritability for number of leaves per plant were 11.67-86.0, 46.67 and 89.28%; and for total leaf area were 142.83-853.50, 42.49 and 88.19%, respectively, as reported by Lal and Haque (1972). In the present study, for leaf potential per plant (no. of leaves/plant), the range recorded at both the locations, Solan and Katrain, was 17.0-55.2 and 40.8-106.2, respectively, indicating the influence of location on the expression of this trait. The coefficient of variation at genotypic level at Solan was about twice that of Katrain and higher heritability to the extent of 45% was also recorded at Solan, which was, however, lower in magnitude than the earlier report (Lal and Haque, 1972). However, lower magnitude of genotypic variance than environmental variance at both the locations, indicated the larger

Table 1. Estimates of mean, range, variances (phenotypic, genotypic, environmental), coefficients of variation (genotypic, phenotypic, environmental) and heritability for leaf and root characteristics

Character	Location	Mean $\pm$ S.E.	Range
Leaf potential/plant	Solan	30.37 $\pm$ 4.12	17.00-55.20
	Katrain	69.45 $\pm$ 9.02	40.80-106.20
Leaf area	Solan	55.87 $\pm$ 6.98	32.435-96.645
	Katrain	44.75 $\pm$ 4.33	29.196-65.183
Nodules/plant	Palampur	133.42 $\pm$ 29.63	64.5-290.0
	Solan	46.44 $\pm$ 18.48	3.00-131.0
Nodules dry weight/ plant	Palampur	0.4192 $\pm$ 0.0885	0.220-0.940
Root dry weight/plant	Palampur	0.8285 $\pm$ 0.1956	0.300-1.825

\*Significant at 5% level.

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Pheno- typic	Variances		Coefficient of variation			
	Genotypic	Environmental	Pheno- typic	Geno- typic	Environ- mental	Herita- bility
62.10	28.16	33.94	25.95	17.47	19.19	45.34*
198.31	35.58	162.73	20.27	8.58	18.36	17.94
136.75	39.26	97.49	20.93	11.22	17.67	28.71*
43.07	5.63	37.44	14.67	5.30	13.67	13.07
1998.43	243.10	1755.33	33.51	11.69	31.40	12.16
835.45	152.12	683.33	62.24	26.56	56.29	18.21
0.01588	0.00021	0.01567	30.07	3.49	29.86	1.35
0.07837	0.00188	0.07649	33.79	5.24	33.38	2.40

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influence of unknown factors. The influence of location was also considerable as the heritability at one location was two and a half times more than the other. For leaf area, mean and range were higher at Solan than those of Katrain. Lower estimates of error variances for this trait also indicated its sensitivity to unknown factors. For this trait, also, the magnitude both genotypic coefficient of variation and heritability estimates at Solan were more than twice those at Katrain, indicating the influence of location on these estimates. In general, a wide range of variation was recorded both for leaf area and leaf potential among genotypes at both the locations, however, the significant differences among genotypes were observed at Solan location.

The information on root traits is also limited. However, Mitchell and Russell (1971) reported a range of 0.4 to 2.0 gm per plant root dry weight, recorded 60 days after planting and presence of varietal differences for this trait. The range recorded in the present study for this trait was from 0.30 to 1.82 gm per plant. The heritability for root dry weight was, however, very low due to high coefficient of variation at environmental level. For nodules dry weight, also, similar situation was observed with respect to range, variation and heritability. Nodules per plant, which were counted at two locations, namely Palampur and Solan, the estimates of mean and range as well as coefficient of variation at genotypic, phenotypic and environmental level and heritability were higher at Solan. However, at both the locations, high magnitude of variance and coefficient of variation at environmental level, indicated the greater influence of environment due to unknown factors, one of them might be sampling error. In general for root characteristics, a wide range among the genotypes studied was recorded, though heritability estimates remained low due to greater influence of unknown factors.

Conclusion. Wide range of variability is present for leaf and root characteristics, which is, however, greatly influenced both by location and some unknown factors and leaf characters had comparatively higher heritability than that of root characteristics. Therefore, it would be possible for a breeder to direct his selection at least for leaf characteristics, leading to higher photosynthetic activity and enhanced yield.

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V. P. Gupta  
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107 <sup>245</sup> Genetic, altitude and climatic effects on seed yield and germinability traits in soybean.

Introduction. In soybean production, the lack of varieties having good seed quality, with respect to germinability, is an important constraint. Though several authors (Johnson et al., 1955; Schutz and Bernard, 1967; Shorter et al., 1977) have reported the role of genotype, environment, and genotype x environmental interaction for seed yield and seed weight, yet such information in respect to seed quality traits, such as percent hard seeds and germinability is meager (Gupta and Garg, 1980). Therefore, in the present study, an attempt has been made to obtain information on the role of genetic, altitude and other climatic factors on seed yield and seed quality traits.

Methods and materials. Sixty varieties of soybean were grown in completely randomized block design with two replications at Palampur, Kangra and Kulu having altitudes of 1300, 700 and 1300 m, respectively, during Kharif, 1973. The average monthly meteorological observations for the growing period of soybean in respect to different locations are given in Table 1. The plot size was 2.7 sq. m. at Kangra and Kulu, while it was 3.0 sq. m. at Palampur. Data were recorded for seed yield (kg/ha), 100-seed weight (g), percent hard seeds and germinability. Days taken to 70% germination was taken as the criterion for germinability. For this purpose, a sample of 100 seeds was kept in the seed germinator at 90°F. The count for the germinated seeds was made after 24 hours. Seeds that did not absorb moisture in the germination test were counted and expressed as percent hard seeds. The combined analysis of variance over locations was done and coefficients of variation, heritability and the expected genetic advance were also calculated. Locations were measured for their potential as per Finlay and Wilkinson (1963) by estimating environmental index for each location.

Results and discussion. Combined analysis of variance (Table 2) over locations indicated significant mean squares due to genotypes, locations, and genotype x location interactions for seed yields, 100-seed weight and percent hard seeds. In case of days to 70% germination, mean squares due to genotypes and locations were only significant, indicating the absence of genotype x location interaction.

On partitioning the total variability, it was found that all the characters except germinability had the highest coefficient of variation for genotype x location interaction (Table 3). The coefficient of variation at genotypic and location level was considerably less for these characters. Among all the seed characters studied, percent hard seeds exhibited the highest genotype and genotypic x location coefficient of variation.

Contribution of each component of variability for the expression of seed yield and other characters is given in Table 4. Genotype x location interaction component has predominantly contributed (66 to 76%) for seed yield and 100-seed weight, whereas, for percent hard seed, its contribution is around 32%. In the present material, genetic component also contributed from 20-30% for seed yield, percent hard seed and germinability. It is interesting to note that for 100-seed weight, genetic component is playing the least role and more than 90% variation has been caused by genotype x location interaction and location components of variability.



Table 1. Average (over 5-10 years) monthly meteorological observations during growing period of soybean at different locations

Meteorological observation	Location	Months				
		June	July	Aug.	Sept.	Oct.
Average mean temperature ( $^{\circ}\text{C}$ )	Palampur	25.7	24.8	24.7	22.9	20.9
	Kangra	26.5	25.5	25.0	24.3	21.3
	Kulu	24.3	23.8	24.0	22.0	19.5
Average minimum temperature ( $^{\circ}\text{C}$ )	Palampur	21.7	22.1	22.1	19.7	17.1
	Kangra	17.5	21.3	21.0	18.8	14.2
	Kulu	18.5	17.7	18.5	15.1	10.6
Average maximum temperature ( $^{\circ}\text{C}$ )	Palampur	29.6	27.5	27.2	26.1	24.6
	Kangra	35.5	29.7	29.0	29.7	28.4
	Kulu	30.1	29.0	29.5	28.9	28.4
Average rainfall (mm)	Palampur	367.2	700.0	800.0	258.6	43.4
	Kangra	136.3	402.3	649.9	141.1	33.5
	Kulu	25.3	60.1	58.2	19.7	10.4
Average relative (%) humidity	Palampur	61	78	85	78	62
	Kangra			Not available		
	Kulu	85	86	85	85	77
Average rainy days (no.)	Palampur	2.0	5.3	6.6	1.9	1.0
	Kangra	7.4	12.3	16.3	6.2	1.5
	Kulu	1.7	4.0	3.6	2.0	0.8

Table 2. Combined analysis of variance over environments

Source	Degrees of freedom	Mean sum of squares due to			
		Seed yield (kg/ha.)	100-seed weight (gm)	Percent hard seed	Days to 70% germination
Genotype	59	466138.4*	23.3*	28.55*	0.77*
Environments	2	13249800.0*	332.5*	103.85*	17.40*
Genotype x Environment	118	205788.6*	23.0*	9.63*	0.19
Pooled Error	177	5033.0	0.88	3.12	0.25

} 0.23

\*Significant at the 5% level.

Table 3. Components of variance and coefficient of variation from combined analysis over locations

Components of variance & C.V.	Character			
	Seed yield (kg/ha.)	100-seed wt. (gm)	% hard seeds	Days to 70% germination
Genotypic variance	86783.27	0.5	3.15	0.09
Locational variance	9326.59	2.58	0.78	0.14
Genotypic x location interaction variance	102869.14	11.06	3.26	--
Error variance	50.33	0.88	3.12	0.23
Genotypic coefficient of variance	13.5	1.6	57.2	7.30
Genotypic x location C.V.	20.8	23.9	58.1	--
Locational C.V.	6.2	11.5	28.5	9.20
Experimental Error C.V.	0.4	6.7	56.9	11.60

Table 4. Percent contribution of each component of variance for the expression of seed yield and its quantity characters

Character	Components of variance			
	$\sigma^2_g$	$\sigma^2_{gl}$	$\sigma^2_{e_l}$	$\sigma^2_e$
Yield (kg/ha)	27.88	66.09	5.99	0.032
100-seed weight (gm)	0.34	75.91	17.70	6.04
Percent hard seeds	30.57	31.56	7.61	30.25
Days to 70% germination	19.44	--	30.89	49.6

Estimates of heritability and genetic advance expressed as percent of mean at different locations and after eliminating locations effects on genotypes are given in Table 5. At each location, a high heritability associated with high genetic advance was noticed for all the traits. The results suggested the presence of additive genetic effects at all the locations. However, after eliminating the location effects on genotypes, the magnitude of both heritability and genetic advance decreased for seed yield, percent hard seeds and days to 70% germination. In case of 100-seed weight, the estimates of heritability and genetic advance were drastically reduced to the lowest level. The results suggested the influence of the locations on these parameters.

In order to quantify the environments, estimates of environmental index were calculated as the deviation of such location mean from grand mean (Table 6). It can be seen that Kangra and Palampur were the best locations for the expression of seed yield, while Palampur appeared to be the best for 100-seed weight. Kulu was the best environment for germinability. The frequency of percent hard seeds was the highest at Kangra. The study of this information reveals that for seed yield apart from altitude, other climatic factors such as humidity, temperature and rainfall also play an important role (Table 1). The low yield of soybean genotypes at Kulu appears to be due to the low mean temperature during the growth period and more specifically after September when the seeds develop. For the expression of higher 100-seed weight, comparatively high rainfall and relatively low maximum temperature at Palampur during seed development appear to be responsible. The lower elevation and comparatively higher maximum temperature at Kangra produce more hard seeds. Low temperature and less rainfall during seed development appear to be responsible for better germinability at Kulu.

Table 5. Estimates of heritability and genetic advance expressed as percent of mean at different locations as well as after eliminating locational effects on genotypes

Character	Estimate of	Kangra	Palampur	Kulu	After eliminating
					location effects on genotypes
Seed yield (kg/ha)	Heritability	72.17	61.21	36.08	29.60
	Genetic advance as % of mean	82.31	54.59	125.91	17.23
100-seed weight (gm)	Heritability	95.15	92.83	84.94	0.40
	Genetic advance as % of mean	35.61	43.47	42.20	2.05
Percent hard seeds	Heritability	84.12	77.62	85.79	33.00
	Genetic advance as % of mean	218.72	199.43	263.23	67.68
Days to 70% germination	Heritability	47.16	58.97	36.08	28.10
	Genetic advance as % of mean	70.72	75.84	51.75	32.73

Table 6. Estimates of environmental index

Character	Environments			Grand mean
	Palampur	Kangra	Kulu	
Seed yield (kg/ha)	+83.7	+87.7	-171.3	1541.3 <u>+ .64</u>
100-seed weight (gm)	+ 2.7	- 1.4	- 1.3	13.9 <u>+ .085</u>
Percent hard seeds	- 0.5	+ 1.5	- 1.0	3.1 <u>+ .14</u>
Days to 70% germination	- 0.3	- 0.3	+ 0.6	4.1 <u>+ .04</u>

Summary. Fifty exotic germplasm lines, along with ten extensively evaluated varieties of soybean, were evaluated in a replicated trial at three locations having altitudes 700 to 1300 m above mean sea level, in Himachal Pradesh (India); for seed yield, seed size, percent hard seeds and germinability to understand the nature and magnitude of variation present for these characters. Significant differences were observed among genotypes for characters studied. Genotype x environment interaction was significant for seed yield, 100-seed weight and percent hard seeds. Environmental influence was also significant for all the traits. Highest C.V. at genotypic, genotype x location and location level was observed for percent hard seeds. Estimates of heritability and genetic advance were influenced by location for all the characters studied. After eliminating the location effect, the highest heritability was recorded for percent hard seeds, followed by seed yield and germinability; and 100-seed weight was the most influenced by environments for genetic parameters. Except germinability, the expression of variation for all other traits was mainly due to genotype x location effects, besides genetic and location. For seed weight, it was mainly genotype x location interaction that accounted for 76% of the total variation. For germinability, the effect of altitude was alone appreciable besides genetic component. For seed yield, Palampur and Kangra were the best environments. For seed weight, Palampur was the best, whereas at Kangra, the frequency of hard seeds was maximum. Kulu Valley with subtemperate climate was best for germinability.

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11) <sup>745</sup> Correlation among seed yield, seed quality and nutritional traits in soybean [ ]

Introduction. In the present communication, the information obtained on correlations among 14 traits related to seed yield, seed quality and nutrition in soybean germplasm has been discussed. The information on the nature of variation for these traits in the above material has been reported earlier (Rana et al., 1981).

Materials and methods. The materials and methods were reported earlier by Rana et al. (1981). Correlation coefficients were, however, estimated among the 14 traits on the basis of unadjusted means of 250 germplasm lines using the standard formula.

Results and discussion. Estimates of correlation coefficients among the 14 characters, namely, seeds per plant, yield per plant, seed weight, 100-seed volume, water absorption, percent germination, percent hard seeds, crushing hardness, percent protein, percent potassium, percent phosphorus, percent moisture, boldness index and specific-gravity index, calculated on the basis of observed mean values of 250 test cultures, are given in Table 1.

Out of the 91 possible combinations among the 14 characters studied, 37 combinations showed significant correlation coefficients.

With respect to seeds per plant, boldness index, specific-gravity index and 100-seed volume, yield per plant has shown strong positive association indicating, thereby, that seed yield in soybean can be improved by making selections on the basis of specific-gravity index, which, in turn, reflects seed weight, seed boldness and 100-seed volume. Interestingly, these traits bear significant positive correlation among themselves. Positive association of seed weight with seed yield in soybean has also been documented earlier (Johnson et al., 1955; Keller et al., 1978). However, a few workers (Kwon and Torrie, 1964; Hartwig and Edward, 1970; Shettar et al., 1978) have reported absence of any association of seed weight with seed yield or its negative association. Correlation of seed number with seed yield has been very well documented earlier (Leffel and Henson, 1961; Gopani and Kabaria, 1970; Jaranowski et al., 1980). With respect to 100-seed volume and specific gravity index some workers have reported absence of any association with seed yield (Smith and Weber, 1968; and Gupta and Garg, 1980).

Among all the major yield contributing seed traits, [breeding] for specific-gravity index, specifically following the stratified method of mass selection would help in simultaneously improving the seed size and seed yield. Interestingly, both specific-gravity index and seed yield are positively associated with water absorption capacity of seed which in the present study has been taken as one of the criteria of good cooking quality -- the higher the water absorption capacity of a genotype, the better the cooking quality of it. Therefore, the present correlation analysis indicates that seed yield, seed size and cooking quality in soybean can be improved simultaneously. Information on the relationship of water absorption with seed yield and other seed quality in the literature is as good as nil. ✓

In the present material, seed yield has not been found to bear any relationship with percent protein, percent potassium, percent phosphorus, percent moisture, percent hard seeds and percent germination. With respect to percent

Table 1. Estimates of correlation coefficient among various characters studied on the basis of 250 test cultures

Characters	1	2	3	4	5	6	7	8	9	10	11	12	13
Yield/plant (gm)	.77*												
Seed wt (mg)	-.16*	.44*											
100-seed volume (cc)	.02	.26*	.38*										
Water absorption	-.23*	.30*	.35*	.35*									
% germination	.26*	.07	-.24*	-.04	-.23*								
% hard seeds	-.03	-.10	-.11	-.07	-.30*	-.24*							
Crushing hardness (kg)	-.20	-.13*	-.01	.08	-.08	-.20	.22*						
% protein	-.03	-.06	-.02	.04	.03	.05	.02	.13*					
% potassium	-.06	-.01	.88	.07	.04	-.05	-.05	-.04	.04				
% phosphorus	.05	-.06	-.11	-.04	-.22*	.13*	.05	-.12*	.04	-.06			
% moisture	.05	.01	-.06	-.05	-.11	-.08	.11	-.14	-.05	-.02	.11		
Boldness index	-.13*	.40*	.86*	.35*	.78*	-.21*	-.08	-.05	-.01	-.01	-.16*	-.05	
Specific gravity index	.12*	.29*	.27*	.14*	.16*	.12*	-.08	-.09	-.01	-.05	-.04	-.01	.39*

\*Significant at the 5% level.

potassium, percent phosphorus, percent moisture and crushing hardness, there is no information available in the literature on their association with yield. Garg (1979) reported percent hard seeds to have no association with yield, while Klykov (1952) found a positive association of grain yield with number of hard seeds. Earlier workers have also reported absence of any relationship between seed yield and percent germination (Singh et al., 1979; Garg, 1979). Another important character investigated in the present study is crushing hardness as an index of cooking quality. It has shown negative association with yield. Though this correlation is significant, yet it accounts for only 1.7% of the total variation in yield. As no information is available in literature on this association, it requires further study.

With respect to associations of percent protein and seed yield, the reports in the literature are conflicting ones, some showing no correlation (Smith, 1967); some giving negative association (Arora et al., 1970; Kwon and Torrie, 1964); and others giving positive relationship (Brim and Burton, 1979). Shannon et al. (1972) have reported positive and negative associations of protein with yield in two different populations. Percent protein has been found to be positively associated with crushing hardness in the present material as also reported earlier by Gupta et al. (1980). However, protein content has not shown any association with percent potassium, percent moisture, boldness index, specific gravity index, seed weight, 100-seed volume, water absorption and number of hard seeds in the present study. Correlation of protein content with the content of phosphorus, potassium, and moisture and capacity of water absorption does not seem to have been studied earlier as there are no reports to this effect in the literature. Seed size, in the literature, has been reported to be positively correlated with high protein content (Fehr and Weber, 1968). Percent protein has been reported to have positive correlation with specific gravity (Hartwig and Collins, 1962; Fehr and Weber, 1968; Kwon et al., 1971) though there is no association between the two in the present material. The probable reason for the contradictory findings in the present material may be the extraordinarily large number of genotypes varying in seed weight, seed volume, and boldness index, which is further confirmed by the positive association of specific-gravity index, with seed weight, seed volume and boldness index recorded in the present material.

Another important character studied is the percent germination, in which, due to high magnitude of block environmental effects, the analysis of variance did not show significant difference among genotypes, yet the wide range of variation observed (18 to 98%) warrants the consideration of this trait in relation to others. Percent germination is positively associated with seeds per plants, but interestingly had no association with yield per plant, 100-seed volume, percent protein, percent potassium and percent moisture. It is interesting to note that germination exhibited positive association with specific gravity index and phosphorus content. Such information was not available in the literature. However, percent germination is negatively associated with boldness index, crushing hardness, percent hard seeds, water absorption and seed weight. With respect to relationship of germinability with crushing hardness, boldness index and water absorption, this is the first report. Negative association between germination percentage (both under laboratory as well as field conditions) and seed size has been widely reported (Edward and Hartwig, 1971; Paschal and Ellis, 1978; Singh et al., 1978). Johnson and Leudders (1974) and Garg (1979) have reported absence

of any associations between seed size and percent germination, while positive correlation between the two parameters has been reported by Burris et al. (1971) and Codey et al. (1974). It appears that, for having high germinability, one should select genotypes having high specific-gravity index, high phosphorus content, on the one hand having low seed weight, low water absorption capacity, less hard seeds, less boldness index and low crushing hardness. Negative association between crushing hardness and percent moisture as well as between phosphorus content and boldness index is being reported for the first time consequent upon the findings of the present investigation.

Conclusion. Correlation analysis indicated that specific gravity index can be used effectively for improving seed yield in soybeans, but for improving seed germinability besides specific-gravity index, one has to breed for high phosphorus content but with lesser hard seeds, low crushing hardness, low water absorption capacity and smaller seed size.

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1) <sup>145</sup> Constraints in using seed leachate characteristics to estimate seed vigor for varietal seed keeping quality comparisons in soybeans,

Summary. Recent research has provided good evidence that characteristics of seed leachate, such as electrical conductivity and optical density, can be used as a measure of relative vigor for different seed lots of the same variety. Because plant breeders selecting for better seed longevity must make comparisons among varieties, studies were conducted to determine if leachate characteristics could be used to compare seed vigor among varieties that vary in seed size, seed color and seed coat permeability.

Seeds of a yellow-seeded variety (TGM 579) and a black-seeded variety (TGM 618) were soaked with seed coats either intact or removed. To mimic varietal differences in seed size, samples of 8, 10, 12 and 14 seeds were used. A second experiment was conducted to determine if varietal differences in hard seededness would confound varietal comparisons in seed vigor. Leachate conductivities of 5 varieties with slow seed imbibition were compared with those from 5 varieties that imbibe rapidly.

These studies showed that seed coat pigments influence optical density values of leachate and that varietal differences in hard seededness influence conductivity. Thus, it appears that for comparisons among varieties, seed coat removal would be necessary. Seed size also influences leachate conductivity values such that seeds would need to be weighed to adjust conductivity scores to remove the influence of seed size. Because seed coat removal is time consuming it seems unlikely that seed leachate analysis will be used as a screening criterion by plant breeders who require rapid tests for comparisons among different genotypes.

Introduction. Soybean seeds have a relatively short storage life and rapid loss of seed vigor is an acute problem in the tropics where high temperatures and high relative humidities hasten seed deterioration. Varieties with genetically superior seed storability have been identified (Wien and Kueneman, 1981), and crosses have been made to incorporate the improved seed longevity into high yielding backgrounds adapted to different tropical regions. Thousands of progenies are being screened at IITA for their inherent storability. Seeds of breeding lines are aged, either by a modified accelerated aging technique (IITA, Annual Report, 1977) or by ambient storage, and subsequently evaluated for their relative vigor by emergence tests from soil.

It was felt that a laboratory test for seed vigor after aging might improve screening precision. Seed leachate characteristics such as electrical conductivity and optical density have shown promise as means to evaluate the relative vigor of different seed lots of the same variety (Matthews and Bradnock, 1968; Parrish and Leopold, 1977; McDonald, 1977; Yaklich and Kulik, 1979). Little information, however, is available on seed factors that might influence the use of leachate characteristics for relative seed vigor evaluation among varieties. Effects of three seed characteristics (seed size, seed coat color and seed coat permeability), thought likely to confound comparison among varieties, were evaluated in this study.

Materials and Methods. Experiment 1: Two soybean varieties that varied in seed coat color but were similar in seed size were chosen for the study. Yellow-seeded TGm 579 and black-seeded TGm 618 had seed sizes of 11.7 and 11.3 g/100 seeds, respectively. Seedling emergence from soil was greater than 80% for both varieties, indicating high seed vigor.

Two lots of 8, 10, 12 and 14 seeds of each variety -- one lot with intact seed coats and the other lot with seed coats removed -- were separately soaked in 20 mls of distilled water in glass vials for 4 hr. The experiment was completely randomized with three replications. After soaking, the solution was decanted into fresh vials for measurement of leachate conductivity ( $\mu$  mho/cm) and optical density at 390 nm.

Experiment 2: This experiment was conducted to determine whether varietal differences in seed coat permeability would confound comparison in seed vigor determined from seed leachate conductivity measurements. Ten IITA germplasm lines were harvested at physiological maturity (when pods were yellow) and kept in a dry environment to eliminate field weathering. After all pods were dry (4 weeks), plants were threshed by hand. From a previous study it was known that TGm 1, TGm 80, TGm 479, TGm 715 and TGm 748 are soft seeded (imbibe water rapidly), while TGm 46, TGm 94, TGm 106, TGm 112 and TGm 920 have a high percentage of hard seed. One hundred seeds of each of the 10 varieties were soaked in 60 mls of distilled water in petri dishes. After 1 hr soaking the number of unimbibed seeds was recorded. The leachate conductivity of the solution in which the seeds were soaked was measured for each of the 10 varieties. This experiment was completely randomized with 2 replications.

Results and Discussion. Effects of seed size: Increasing the number of seeds soaked from 8 to 14 increased the electrical conductivity and optical density of leachate (Tables 1a and 1b). This suggests that varietal differences in seed size could confound varietal comparisons for seed vigor based on leachate characteristics. This is in contrast with results of Tao (1978) who did not find significant differences in leachates conductivity among seed size groups within a variety. As in our study, Yaklich et al. (1979) found seed size to have a measurable effect on leachate conductivity. They noted, and we concur, that the effect of seed size can be minimized by reporting leachate conductivity in  $\mu$  mhos/cm/g seed.

Effects of seed coat pigments: Leachate electrical conductivity values increased similarly for both varieties when seed coats were removed (Table 2a), suggesting that pigments do not influence conductivity scores and, therefore, it would not be necessary to remove seed coats when comparing storability of varieties with different seed coat colors. In contrast, seed coat removal did result in a markedly different response for the 2 varieties in regard to optical density of leachate (Table 2b). Optical density values were higher for black-seeded TGm 618 than for TGm 579 when coats were left intact, but were higher for TGm 579 when coats were removed. Consequently, it would be necessary to remove seed coats for varietal comparisons of seed vigor based on leachate optical density values. Seed coat removal is a slow process and, therefore, not practical for breeders who need to evaluate large numbers of progenies. Optical density measurements of leachate may, however, be a useful measure of seed deterioration for seed physiologists (Priestly and Leopold, 1980) when only relatively few samples are to be tested.

Effects of differential seed coat permeability: Five varieties with high percentage of hard seeds had significantly lower leachate conductivity scores than the five varieties with a low percentage of hard seeds (Fig. 1), suggesting that varietal differences in seed coat permeability could confound varietal comparisons for seed vigor based on conductivity scores. For breeding purposes, it would be necessary to remove seed coats for varietal comparisons.

Conclusions. Seed leachate characteristics such as electrical conductivity and optical density do not readily lend themselves for use as a screening method for making varietal comparisons for seed keeping quality because seed coats would necessarily have to be removed. Further, adjustments in leachate conductivity or optical density scores would need to be made for varietal differences in seed size. Replicated seedling emergence tests from uniform seed bed appear to be a better method for relative seed vigor evaluations among varieties.

Table 1a. Effect of varieties and seed number on leachate conductivity ( $\mu$  mhos/cm) of TGm 579 and TGm 618

Varieties	Number of seeds soaked				Mean	L.S.D. (.05)
	8	10	12	14		
TGm 579	157.5	267.3	304.0	298.7	256.9	32.67
TGm 618	115.8	138.8	190.2	241.2	171.5	
Mean	136.7	203.1	247.1	269.9		
L.S.D. (.05)	46.2					
Mean <sup>a</sup>	17.08	20.3	20.6	19.3		

<sup>a</sup>Means after dividing the conductivity score by the number of seeds soaked to remove the effect of seed size.

Table 1b. Effect of varieties and the number of seed soaked on leachate optical density at 390 nm, of TGm 579 and TGm 618

Varieties	Number of seeds soaked				Mean	L.S.D. (.05)
	8	10	12	14		
TGm 579	0.089	0.128	0.154	0.177	0.137	0.108
TGm 618	0.173	0.214	0.252	0.307	0.237	
Mean	0.131	0.171	0.203	0.242		
L.S.D. (.05)	0.153					
Mean <sup>a</sup>	0.016	0.017	0.017	0.017		

<sup>a</sup>Means after dividing the optical density score by number of seeds soaked to remove the effect of seed size.

Table 2a. Effect of varieties and seed coat on leachate conductivity ( $\mu$  mhos/cm) of TGm 579 and TGm 618

Varieties	With seed coat	Without seed coat	Mean	L.S.D. (.05)
TGm 579	200.2	313.6	256.9	32.67
TGm 618	135.8	207.2	171.5	
Mean	168.0	260.4		
L.S.D. (.05)	32.67			

Table 2b. Effect of varieties and seed coat on leachate optical density at 390 nm of TGm 579 and TGm 618

Varieties	With seed coat	Without seed coat	Mean	L.S.D. (.05)
TGm 579	0.025	0.249	0.137	0.108
TGm 618	0.339	0.134	0.237	
Mean	0.182	0.192		
L.S.D. (.05)	0.108			

Table 3. Leachate conductivity of 5 soybean varieties with high percentage of hardseed and 5 varieties with low percentage of hardseed

Variety	% hardseed	Conductivity (m ho/cm)
High percentage of hardseed		
TGm 94	31	85
TGm 112	32	75
TGm 46	38	61
TGm 106	43	128
TGm 920	44	108
Low percentage of hardseed		
TGm 479	0	250
TGm 715	2	148
TGm 80	2	199
TGm 748	7	307
TGm 1	12	212



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17 Relationship between photoperiod, temperature, solar radiation and grain yield in soybean.

Photoperiod and temperature influence the growth, development and area of adaptation of soybean cultivars (Shanmugasundaram, 1981). Germplasm collected from the U.S. Maturity Groups 00 to X were screened for photoperiodic response and were classified into 10 different photoperiod sensitivity score groups, based on flowering in 10 and 16 hr photoperiods (Shanmugasundaram, 1981). By utilizing 0 sensitivity score types as one parent, high yielding selections with different levels of photoperiod sensitivity have been developed.

A total of 61 and 39 selections in 1980 and 1981, respectively, was evaluated for yield in spring, summer and autumn in advanced yield trials.

Using the natural day length and controlled light rooms either to extend day length or to provide darkness, days-to-flowering were determined for each selection in 10, 12, 14 and 16 hr photoperiods. The deviation in days-to-flowering between 10 and 16 hr (A), 10 and 14 hr (B), 10 and 12 hr (C), 12 and 14 hr (D), 14 and 16 hr (E) and 12 and 16 hr (F) were determined and a photoperiod sensitivity score was assigned to each selection for each of the above categories A to F.

The degree-day was computed for each selection by summing the mean daily temperature above 15°C from sowing to maturity of each selection (Nuttonson, 1957). Similarly, solar radiation required from sowing to maturity for each selection was determined by summing the daily solar radiation from sowing to maturity. Simple correlation coefficients between grain yield, photoperiod sensitivity scores, the degree-day and the solar radiation were determined.

Two years' results suggest that the relationship between grain yield and the other environmental variables varied with the season. None of the variables had any influence on grain yield in the autumn season. During the summer season, however, low photoperiod scores (i.e., insensitive selections) were correlated with higher yields and vice versa. Results from the spring season suggest that sensitivity to photoperiod is desirable to obtain high yield (Table 1). Similarly, there was a negative correlation between degree-day and solar radiation and high yield for the summer season, only suggesting that higher degree-days or solar radiation lowers yield and vice versa (Table 2). No significant relationship between degree-day and yield was evident during the spring or autumn seasons. However, yield was positively correlated with solar radiation during the spring season (Table 2).

Our results suggest the advisability of selecting photoperiod insensitive lines with high yield potential at least for latitude 23°N to successfully produce high yielding selections in the summer season. During early spring and late autumn the temperatures at our location are much lower than during other times. We speculate that either the temperature or the photoperiod X temperature interaction during spring and autumn exert more influence on yield than does photoperiod alone. Therefore, temperature response of the photoperiod insensitive selections need to be investigated.

Table 1. Simple correlation coefficient between yield and photoperiod sensitivity score in three seasons during 1980 and 1981

Season	Year	Photoperiod sensitivity score					
		A 10-16 hr	B 10-14 hr	C 10-12 hr	D 12-14 hr	E 14-16 hr	F 12-16 hr
Spring	1980	0.34**	0.38**	0.36**	0.45**	NS <sup>a</sup>	0.43**
	1981	0.37*	NS	NS	NS	NS	0.44**
Summer	1980	-0.54**	-0.45*	-0.27**	-0.45**	-0.55**	-0.53**
	1981	-0.33**	NS	-0.36**	-0.33*	-0.34*	-0.35*
Autumn	1980	NS	NS	NS	NS	NS	NS
	1981	NS	NS	NS	NS	NS	NS

<sup>a</sup>NS = Not Significant

\*Significant at the 5% level.

\*\*Significant at the 1% level.

Table 2. Simple correlation coefficient between high yields and either degree-day or solar radiation in three seasons during 1981

Variable	Yield		
	Spring	Summer	Autumn
Degree-day	0.30 NS <sup>a</sup>	-0.32*	0.06 NS
Solar radiation	0.32*	-0.33*	0.07 NS

<sup>a</sup>NS = Not Significant.

\*Significant at the 5% level.

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245  
2) Screening for immature green soybeans as a vegetable [ ].

Soybeans, either immature green beans in pod as in Japan, or shelled immature green beans as in China, are used as a vegetable in a number of countries. Although there is no strict definition for vegetable soybeans, generally large pods and seeds are preferred. The pubescence and hilum color should be gray. Majority of the pods should be either 2 or 3 seeded. The immature green beans cook easily and have mild nutty flavor (Kline, 1980). Countries like Japan have rigid taste and flavor requirements.

We evaluated 136 large-seeded germplasm accessions against two locally grown vegetable soybeans. The experiment was planted in the field on July 2, 1981, in a randomized complete block design with two replications.

The results suggest that the germplasm has both earlier and later flowering and maturing types than the checks. One-hundred-seed weight ranged from 12 to 45 gms with a mean of 24 gm on 13% moisture basis. Percent of pods with two or more seeds ranged from 42 to 100 with a mean of 89% (Table 1).

Among the 136 entries, 63 had gray pubescence, with yellow or light-green seed coat. However, only three accessions had both gray pubescence and 100-seed weight of 30 gm or more, comparable to the check 'Tzurunoku' (Table 2).

The length and width of pods were measured at  $R_6$  stage (Fehr and Caviness, 1977) and were related to the 100-seed weight. Our results show that even among the germplasm, pod width and pod length are highly significantly associated with 100-seed weight ( $r = 0.66^{**}$  for both pod length and pod width with 100-seed weight of 2-seeded pods and  $r = 0.63^{**}$  for 3-seeded pods). Frank and Fehr (1981) reported that pod length and pod width were significantly associated with 100-seed weight and can be used as an indirect selection criteria for 100-seed weight.

Sample seeds of the four large-seeded soybeans (Table 3) will be available for trials.

Table 1. Variability for time to flowering and time to maturity of 136 large-seeded soybeans compared with two local vegetable types

	Days to			100-seed	%
	$R_1^a$	$R_6$	$R_8$	wt (g) <sup>b</sup>	2-seeded pods
For 136 germplasm					
Range	20-47	56-113	71-129	12-45	42-100
Mean	33	81	95	24	89
CV %	16	12	10	24	8
Tzurunoku (CK)	33	83		30	95
Zen Wu #2 (CK)	26	75		12	90

<sup>a</sup>Fehr and Caviness (1977).

<sup>b</sup>On 13% moisture basis.



Table 2. Classification 136 large-seeded soybeans based on seed coat color, pubescence color and 100-seed weight

Seed coat color	100-seed weight <sup>a</sup> (g)	With acceptable <sup>b</sup> pubescence			With tawny pubescence		
		<19.9	20.0-29.9	>30.0	<19.9	20.0-29.9	>30.0
Yellow and light green	10	50	3	11	28	4	
Black	0	1	3	0	12	5	
Brown	0	0	0	0	4	1	
Mottled	0	0	0	1	3	0	

<sup>a</sup>On 13% moisture basis.<sup>b</sup>Gray or light colored and sparse.

Table 3. Agronomic characters of 3 vegetable types with yellow or green seed coat and gray pubescence selected from 136 large-seeded germ-plasm

AVRDC No.	Name or PI number	100 seed wt <sup>a</sup> (g)	— Days to —			cm				% of 2 or more seeded pods
			R <sub>1</sub>	R <sub>6</sub>	R <sub>8</sub>	2-seed pod L <sup>b</sup>	2-seed pod W <sup>b</sup>	3-seed pod L	3-seed pod W	
G 10134	Green light	34	34	83	92	5.1	1.4	5.8	1.3	73
G 7321	157424	33	35	82	106	4.7	1.3	5.5	1.3	99
G 10157	Chou Hou	31	33	80	100	5.0	1.2	5.9	1.3	92
G 9053	Tzurunoku (CK)	30	33	83	101	5.0	1.2	5.9	1.2	95

<sup>a</sup>On 13% moisture basis.<sup>b</sup>L: Length, W: Width.

### Acknowledgment

We thank Drs. R. L. Bernard and E. E. Hartwig for providing part of the germplasm.

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Yield evaluation of immature, green soybeans [ ].

Soybeans are commonly called the "meat of the fields" in the orient. Soybean is a popular vegetable in oriental diet and is rich in vitamins A and B and is a good source of protein, fat, calcium, phosphorous and iron. Soybean as a vegetable can be grown easily even during the rainy season since the final product to be harvested is immature green seed.

At the Asian Vegetable Research and Development Center one of our objectives is to develop productive vegetable-type soybeans. We evaluated six selections and one accession (Acc.) and two local cultivars in the spring (planted Jan. 31, '81) and autumn seasons (planted Sep. 16, '81). The trial was conducted in a randomized complete block design with four replications.

In Taiwan the farmers are paid based on total plant weight, the middle-men are paid by the companies based on total amount of two- or more-seeded pods. The local market deals with shelled immature green beans. Therefore, data were collected on all the above parameters.

Our results showed that, regardless of the season, some of our breeding lines have the potential to produce a total biomass of 24 t/ha on fresh weight basis and up to 43% of which can be pods. Two- or more-seeded pods ranged from 71 to 92% in the spring and 84 to 92% in the autumn season (Table 1).

AGS 164 yielded 10.7 t/ha of green beans in 94 days in the spring season. Acc. G 8285 in the autumn season produced 17 t/ha green bean in 80 days which is 41% higher yield than the best check, Acc. G 9053. In addition, three of our new breeding lines outyielded the best check in green-bean yield in the spring planting (Table 2).

In an organoleptic test of the vegetable soybeans the desirable selections identified by the Chinese, Indians, Indonesians, Koreans, Filipinos and Americans were G 8285, G 9053, and G 9948. However, Americans and Koreans liked all the entries.

Sample seeds of any of the entries mentioned in this paper may be obtained by writing to the senior author.

Table 1. Total biomass production and their partitioning into component plant parts in vegetable soybeans<sup>a</sup>

AVRDC No.	Total plant wt. t/ha	Leaf	Stem	Pod	Percent of two- or more- seeded pods
<u>Spring season:</u> Date planted - Jan. 31, 1981					
AGS 163	23 ab <sup>b</sup>	7.0 (30) <sup>c</sup>	7.5 (32)	8.7 (38)	86
AGS 164	22 ab	5.0 (23)	8.0 (36)	9.1 (41)	87
AGS 165	22 ab	6.7 (31)	6.8 (31)	8.2 (38)	87
AGS 166	23 ab	6.5 (28)	8.3 (36)	8.5 (36)	92
AGS 167	24 a	5.9 (24)	9.0 (37)	9.4 (39)	88
AGS 168	22 ab	5.1 (23)	7.7 (36)	8.8 (41)	89
G 8285	21 b	5.8 (28)	5.6 (27)	9.5 (45)	86
-----					
G 9053 (CK)	14 c	4.1 (30)	3.2 (24)	6.2 (46)	71
G 9948 (CK)	13 c	3.9 (29)	3.2 (24)	6.2 (47)	87
<u>Autumn season:</u> Date planted - Sep. 16, 1981					
AGS 163	24 a	6.8 (28)	7.1 (29)	10.3 (43)	92
AGS 164	24 a	8.3 (35)	8.5 (35)	7.2 (30)	85
AGS 165	23 ab	7.5 (34)	6.6 (29)	8.4 (37)	89
AGS 166	23 ab	6.2 (27)	8.2 (35)	9.0 (38)	87
AGS 167	22 ab	5.6 (25)	7.7 (35)	9.0 (40)	86
AGS 168	21 b	5.1 (24)	7.7 (36)	8.5 (40)	84
G 8285	20 bc	4.3 (21)	6.3 (32)	9.3 (47)	84
-----					
G 9053 (CK)	23 ab	7.0 (30)	5.7 (25)	10.2 (45)	84
G 9948 (CK)	18 c	5.6 (31)	5.0 (28)	7.5 (41)	91

<sup>a</sup>Data are on fresh weight basis.

<sup>b</sup>Means followed by the same letters are not significantly different at 5% level as per DMRT.

<sup>c</sup>Values in parentheses are percent of total plant weight on fresh weight basis.

Table 2. Immature green-bean yield and selected agronomic characters of 7 vegetable soybean selections

AVRDC No.	Green bean yield <sup>a</sup> t/ha		Days to R <sub>6</sub>		100-seed wt. at R <sub>6</sub>	
	Spring	Autumn	Spring	Autumn	Spring	Autumn
AGS 163	7.8 bc <sup>b</sup>	10.8 b	90	73	31 e	39 de
AGS 164	10.7 a	7.6 b	94	73	41 cd	43 d
AGS 165	7.9 bc	7.3 b	90	66	40 cd	38 e
AGS 166	8.6 b	9.9 b	95	80	41 cd	47 c
AGS 167	9.1 ab	9.7 b	94	80	39 d	39 de
AGS 168	7.3 c	9.8 b	95	80	40 cd	39 de
G 8285	7.4 bc	17.0 a	90	80	51 ab	54 b
G 9053 (CK)	4.8 d	10.1 b	80	73	46 bc	56 b
G 9948 (CK)	6.1 cd	6.8 b	80	66	56 a	64 a

<sup>a</sup>Data are on 90% moisture basis.

<sup>b</sup>Means followed by the same letters are not significantly different at 5% level as per DMRT.

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#### 245 4) Forcing soybeans to mature by spraying paraquat,

In the tropics, soybeans theoretically can be grown year around. The rainfall pattern distinguishes the soybean crop as either wet season or dry season. Rainfall at harvest time invariably causes sprouting of the seeds in the field and can result in a total loss of the crop. However, if a crop can be grown successfully, a season can be saved in the breeding program. An experiment was conducted to artificially force maturity prior to seasonal rains at maturity.

Paraquat at 3.5 liters of ICI commercial product per hectare (0.84 liter a.i. of 1-1' Dimethyl 4, 4' bipyridylum dichloride) was sprayed at either R<sub>6</sub>, R<sub>6.5</sub> or R<sub>7</sub> (Fehr and Caviness, 1977) versus natural maturity without any spray as a control. The experiment was conducted during the summer rainy season using three AVRDC soybean selections: AGS 2, AGS 17 and G 2261.

The results suggest that the soybean crop can be forced to mature by as much as two weeks earlier than normal (control) with paraquat spray at stage R<sub>6</sub> (Table 1). When paraquat was sprayed at R<sub>6</sub> or R<sub>6.5</sub>, the 100-seed weight was reduced by 20 to 34% compared with the control (Table 2). There was a significant yield reduction in treatments with paraquat spray at stage R<sub>6</sub> and R<sub>6.5</sub> as compared with R<sub>7</sub> and control (Table 3).

Table 1. Days from sowing to maturity influenced by paraquat spray at different growth stages in three selections

AVRDC No.	Growth stage when paraquat was sprayed			Control	Mean <sup>a</sup>
	R <sub>6</sub>	R <sub>6.5</sub>	R <sub>7</sub>		
AGS 2	106	106	114	121	112 a
AGS 17	98	98	102	102	100 b
G 2261	88	92	98	102	95 c
Mean <sup>a</sup>	97 d	98 c	104 b	108 a	

<sup>a</sup>Means followed by the same letter are not significantly different at the 5% level as per DMRT.

Table 2. Influence of paraquat spray on 100-seed weight (gm) in three selections

AVRDC No.	Growth stage when paraquat was sprayed			Control	Mean <sup>a</sup>
	R <sub>6</sub>	R <sub>6.5</sub>	R <sub>7</sub>		
AGS 2	4.8	4.8	6.1	6.4	5.5 c
AGS 17	9.9	11.6	12.5	14.9	12.2 b
G 2261	12.3	12.4	14.7	15.4	13.7 a
Mean <sup>a</sup>	9.0 c	9.6 c	11.1 b	12.2 a	

<sup>a</sup>Means followed by the same letters are not significantly different at the 5% level as per DMRT.

Table 3. The grain yield (kg/ha) influenced by paraquat spray at different growth stages in three selections

AVRDC No.	Growth stage when paraquat was sprayed			Control	Mean <sup>a</sup>
	R <sub>6</sub>	R <sub>6.5</sub>	R <sub>7</sub>		
AGS 2	1,350	1,384	2,108	2,211	1,764 a
AGS 17	895	1,324	1,516	1,860	1,399 ab
G 2261	991	938	1,282	1,333	1,137 b
Mean <sup>a</sup>	1,079 b	1,216 b	1,636 a	1,802 a	

<sup>a</sup>Means followed by the same letters are not significantly different at the 5% level as per DMRT.



Table 4. Percent germination of seeds harvested with and without paraquat spray in three cultivars

AVRDC No.	Growth stage when paraquat was sprayed			Control	Mean <sup>a</sup>
	R <sub>6</sub>	R <sub>6.5</sub>	R <sub>7</sub>		
AGS 2	63 (92) <sup>b</sup>	56 (92)	64 (94)	62 (90)	61 a (92 a)
AGS 17	41 (80)	26 (50)	18 (87)	82 (96)	42 b (78 ab)
G 2261	43 (85)	37 (86)	17 (59)	26 (57)	31 b (72 ab)
Mean <sup>a</sup>	49 ab (86) <sup>c</sup>	40 ab (76)	33 b (80)	57 a (81)	

NOTE: There is significant interaction between selections used and the treatments.

<sup>a</sup>Means followed by the same letters are not significantly different at the 5% level as per DMRT.

<sup>b</sup>Open values are without captan seed treatment and values in parentheses are with captan seed treatment.

<sup>c</sup>With captan seed treatment differences were not significant.

The yield reduction of 33 to 40% in treatments of paraquat spray at R<sub>6</sub> and R<sub>6.5</sub> against the control appears to be due to the reduced seed size in the treated plots as opposed to a total loss if rains occur at maturity. However, under the present experimental conditions, the control had significantly higher yield than treatments due to their escape from rain.

Since viability of the harvested seed is important in breeding, a germination test was conducted. The paraquat spray treatments at stage R<sub>6</sub> and R<sub>6.5</sub> did not adversely affect germination compared with the untreated control (Table 4). However, there was significant interaction between selections used and the treatments. If the harvested seeds were treated with captan the percent of germination was markedly improved and there was no significant difference in percent of germination of the seeds in treated and control. Although preliminary results suggest that paraquat spray prior to maturity can hasten maturity and permit soybean production, the significant interaction between selections used and the paraquat spray treatments requires further evaluation before the paraquat spray can be recommended for use either for plant breeding or commercial soybean production in wet season.

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1) A second report on induced mutations for soybean rust resistance\*

Mutation experiments for inducing rust resistance in 11 soybean cultivars have been carried out since 1979. A first part of the results was already published in this Newsletter (Smutkupt et al., 1981). This second paper reports on results of screening and selecting soybean lines for rust resistance.

Seeds of  $M_2$ -bulk,  $M_3$ -bulk and  $M_3$ -single soybeans were grown at Nong Hoi in the rainy season of 1980. Based upon the IWGSR rust rating system, 121 plants with low rust rating were selected from a total of 31,636 plants. Each plant was separately and carefully threshed. No seeds were obtained from selections BM98 and G8587. In the dry season of 1981 only 119 selections were increased in progeny rows in front of the Rangsi Building on Bangkhen campus. Seeds of certain selections did not germinate. In some rows the plants did not grow well and produced no seeds. After harvest, good seeds were obtained from only 90 selections.

All 90 selections were screened for rust resistance in Nong Hoi Valley (altitude about 1000 m above sea level) in Chiang Mai Province (latitude  $18^{\circ}31'$ - $19^{\circ}N$ ) in the rainy season of 1981. Thirty seeds of each selection were planted in 1.50 m-row plot. Fifty six, 28, 28 and 28 rows of four check cultivars (S.J.1, S.J.2, S.J.4 and T.K.5) were planted at the same time.

Seeds of selection line Nos. 81-1-085 (derived from G8586) and 81-1-119 (derived from G8587) did not germinate. Only 88 lines of  $M_4$  and  $M_5$  selections survived for rust evaluation.

The severity of rust disease in Nong Hoi Valley was the same as in the rainy season of 1980. The ratings were made at three different times, 61, 77, and 86 days after planting.

It was observed that the rust disease developed slowly on selections. At 77 days after planting, 82 selection lines/rows were rated 333, 323 and 223-243 in comparison with 87 out of 137 rows of control S.J.1, S.J.2, S.J.4 and T.K.5 which had already reached 343. Most of the selections reached the disease level 343 86 days after planting. The summarized results are shown in Table 1. However, six lines derived from G8586 were still rated 333. In addition, a plant having slow growth of rust (323) from Taichung N No. 81-1-032 was selected.

All rows of 88 selections were harvested. The number of plants in each row was recorded. Check rows of S.J.1, S.J.2, S.J.4 (except T.K.5) were harvested at random. Good seed yield per plant (g), percentage of shrivelled seeds and weight of 100 seeds (g) of all selections and harvested checks were determined. The data of their means  $\pm$  S.E. are shown in Table 2.

Among check cultivars, S.J.1 is a very susceptible check, having the lowest seed yield per plant. In comparing seed yield of selections and checks in S.J.2 and S.J.4 cultivars, the selections of both cultivars show

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Table 1. IWGSR rust reaction on soybean selections rated at three different intervals (days after planting), Nong Hoi Valley in the rainy season of 1981

Cultivars	Number of soybean rows (lines)										
	Total rows	61 days			77 days				86 days		
		IWGSR row-rating in 1981									
		111	123	133, 223	223, 233, 243	323, 333	343	243, 323	333	343	
G 8375	2	0	2 <sup>+</sup> (100) <sup>‡</sup>	0	0	2(100)	0	0	0	2(100)	
Wakashima mutant #10	8	0	7(88)	1(12)	0	8(100)	0	0	0	8(100)	
Taichung N	13	4(31)	6(46)	3(23)	1(8)	9(69)	3(23)	0	0	13(100)	
S.J. 2	4	0	3(75)	1(25)	0	4(100)	0	0	0	4(100)	
S.J. 4	11	2(18)	9(82)	0	0	10(91)	1(9)	0	0	11(100)	
BM 50	4	1(25)	3(75)	0	0	4(100)	0	0	0	4(100)	
BM 98	6	0	6(100)	0	0	5(83)	1(17)	0	0	6(100)	
G 8377	8	0	8(100)	0	1(12)	6(76)	1(12)	0	0	8(100)	
G 8586	23	16(70)	7(30)	0	19(83)	4(17)	0	0	6(26)	17(74)	
G 8587	9	4(44)	5(56)	0	2(22)	7(68)	0	0	0	9(100)	
Total	88	27(31)	56(64)	5(5)	23(27)	59(66)	6(7)	0	6(7)	82(93)	
<u>Check cultivars</u>											
S.J. 1	56	0	47(84)	9(16)	0	5(9)	51(91)	0	0	56(100)	
S.J. 2	28	0	21(75)	7(25)	0	14(50)	14(50)	0	0	28(100)	
S.J. 4	28	2(7)	21(75)	5(18)	0	18(64)	10(36)	0	0	28(100)	
T.K. 5	25	5(20)	16(64)	4(16)	0	13(52)	12(48)	0	0	25(100)	
Total	137	7(5)	105(77)	25(18)	0	50(36)	87(64)	0	0	137(100)	

<sup>+</sup> Number of rows or lines.

<sup>‡</sup>Percentage.

Table 2. Means ( $\pm$  S.E.) of seed weight/plant, shrivelled seeds and weight of 100 seeds of  $M_4$ ,  $M_5$  selections (lines) and check cultivars grown in Nong Hoi Valley in the rainy season of 1981

Cultivars	No. lines evaluated	No. plants harvested	Good seed wt/ plant (g)	Shrivelled seeds (%)	Wt/100 seeds (g)
G 8375	2	7.50 $\pm$ 3.50	0.190 $\pm$ 0.160	67.57 $\pm$ 12.43	11.86 $\pm$ 0.86
Wakashima mutant #10	8	10.88 $\pm$ 3.55	0.607 $\pm$ 0.344	22.23 $\pm$ 13.40	11.89 $\pm$ 0.92
Taichung N	13	13.15 $\pm$ 4.70	0.801 $\pm$ 0.491	33.66 $\pm$ 23.77	12.21 $\pm$ 2.11
S.J. 2	4	17.00 $\pm$ 3.93	0.733 $\pm$ 0.318	22.46 $\pm$ 13.66	7.96 $\pm$ 1.30
S.J. 4	11	13.18 $\pm$ 3.45	1.100 $\pm$ 1.100	19.64 $\pm$ 11.87	11.91 $\pm$ 0.91
BM 50	4	7.00 $\pm$ 2.91	0.816 $\pm$ 0.353	25.91 $\pm$ 4.01	9.44 $\pm$ 1.00
BM 98	6	11.33 $\pm$ 2.28	1.185 $\pm$ 0.606	30.72 $\pm$ 12.43	8.96 $\pm$ 0.72
G 8377	8	14.50 $\pm$ 8.36	1.009 $\pm$ 0.656	17.06 $\pm$ 11.20	7.46 $\pm$ 0.88
G 8586	23	12.65 $\pm$ 7.01	1.431 $\pm$ 1.008	14.51 $\pm$ 5.21	9.17 $\pm$ 0.81
G 8587	9	12.00 $\pm$ 5.43	2.083 $\pm$ 1.051	12.90 $\pm$ 5.19	10.94 $\pm$ 1.20
<u>Check cultivars</u>					
		No. rows evaluated			
S.J. 1	9	13.22 $\pm$ 3.04	0.419 $\pm$ 0.252	28.10 $\pm$ 6.25	8.35 $\pm$ 0.84
S.J. 2	10	13.80 $\pm$ 2.85	0.729 $\pm$ 0.496	15.41 $\pm$ 6.76	8.05 $\pm$ 1.47
S.J. 4	7	12.57 $\pm$ 0.90	0.917 $\pm$ 0.485	17.08 $\pm$ 9.15	9.59 $\pm$ 0.94
T.K. 5	22	3.22 $\pm$ 1.75	2.809 $\pm$ 1.955	24.56 $\pm$ 15.73	12.97 $\pm$ 1.11



Table 3. Certain characteristics of 16 selected selections

Cultivar	Line number	Dose (krad)	IWGSR plant-rating in 1980	IWGSR row-rating in 1981, days after planting			No. plants harvested	Good seed wt/plant (g)	Shrivelled seeds (%)	Wt/100 seeds (g)
Wakashima mutant #10	81-1-013	30	333	123	323	343	13	1.254	14.53	12.030
Taichung N	81-1-029	15	333	123	323	343	11	1.294	14.32	11.840
Taichung N	81-1-031	15	333	123	323	343	20	1.162	14.52	11.220
Taichung N	81-1-032	30	333	111	243	343	17	1.759	8.84	10.750
S.J. 2	81-1-036	30	333	123	323	343	16	1.104	7.92	7.150
S.J. 4	81-1-037	15	323	111	323	343	13	3.479	5.08	12.600
S.J. 4	81-1-038	15	333	111	323	343	16	2.660	6.44	12.660
G 8377	81-1-066	30	343	123	343	343	25	1.888	8.61	7.160
G 8586	81-1-072	15	333	123	243	343	19	4.098	7.74	9.570
G 8586	81-1-078	30	323	123	323	343	24	1.708	7.34	9.380
G 8586	81-1-102	30	323	111	233	343	22	1.179	13.36	10.140
G 8586	81-1-105	30	323	123	323	343	24	2.136	10.80	10.140
G 8586	81-1-107	30	323	111	223	343	21	1.721	10.67	9.530
G 8587	81-1-112	30	333	123	323	343	20	1.292	10.74	12.150
G 8587	81-1-113	15	323	111	323	343	12	2.540	6.50	12.480
G 8587	81-1-114	15	323	123	323	343	17	1.491	8.94	9.240

a better average seed yield per plant than that of the checks. It can be observed in general that the percentage of shrivelled seeds of both selections and checks exposed to severe rust disease is relatively high except in G8586 and G8587.

Based upon a high seed yield per plant with a low percentage of shrivelled seeds, 16 selections derived from Wakashima mutant number 10, Taichung N, S.J.2, S.J.4, G8377, G8586 and G8587 were selected. Data on certain characteristics are shown in Table 3. Their seeds are being increased. These selections will be further tested for yield in the rainy season of 1982.

Soybean rust is still an important disease. Its occurrence is not only found in Southeast Asian countries, but it was likely to occur in Brazil (Chaves and do Vale, 1981). Maiti et al. (1981) reported the reappearance of rust disease in the soybean-growing areas of the northeastern hills of India. A reappraisal of this disease in India has started.

It is evident that a search for additional sources of soybean rust resistance must be continued. The basic biology of this disease must be thoroughly studied. There is also an urgent need to identify the resistance mechanisms.

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<sup>145</sup>  
 1) The effects of temperature on longevity and vitality of soybean seeds [ ]

Studies involving longevity and vitality of seeds after subjection to certain environmental conditions are both numerous and include many species of seeds. When stored under laboratory room conditions, year-to-year plantings are required to maintain adequate quantities of viable soybean seeds for the basic genetics laboratories. Begun in 1971, this study was undertaken for the purpose of investigating the role of temperature in maintaining high germination percentages in seeds subjected to an extended period of storage.

The seeds comprising the lot from which the individual samples were drawn were the progeny of self-pollinated plants whose leaf phenotype was "light" green in color. Produced on the writer's test plot, harvesting of the seeds was delayed until all of the leaves had fallen from the plants and the pods were uniformly dark brown to black in color. Harvested in October, the pods were further air dried by placing a thin layer on a well-ventilated shelf and observing them frequently for one month. The lot was next divided into three samples of approximately 1,000 seeds each. Identified as samples A, B, and C, respectively, each sample was prepared and stored under the following conditions:

<u>Sample</u>	<u>Storage conditions*</u>
A	Laboratory room temperature
B	Household refrigerator
C	Laboratory freezer

\*All samples were placed in plastic containers prior to storage.

Once each year, 50 seeds were drawn from each of the designated samples and tested for longevity and vitality. Testing was accomplished by placing 8 cm of sterilized potting medium (1 part garden soil, 1 part coarse builder's sand, and 1 part vermiculite) into a standard greenhouse propagation flat (35 cm wide, 52 cm long, and 10 cm high). The medium was next firmed, the 50 seeds were spaced uniformly over the surface, and covered with about 2 cm of potting medium. The surface was next firmed, the medium was sprayed until wet throughout, and the flats were placed on greenhouse benches and observed daily for the duration of the testing period. Testing for longevity and vitality was terminated on the 14th day after the seeds were planted.

As may be observed from Figure 1, there is significant fluctuation in germination percentages when individual samples are compared. Seeds stored in the freezer (Sample C) showed 95% germination following a period of ten years in storage. The drastic drop in germination percentage from 82% in 1974 to 58% in 1975 cannot be readily explained. A contributing factor to the low percentage of germination could be the time of the year in which testing was conducted. Records show that this particular sample was tested in late February, while the remaining tests were conducted between late April and early July of each year. While showing less year-to-year fluctuation in germination percentage than Sample C, seeds in Sample B showed 87% longevity following a 10-year period in storage. It should be noted that Sample A

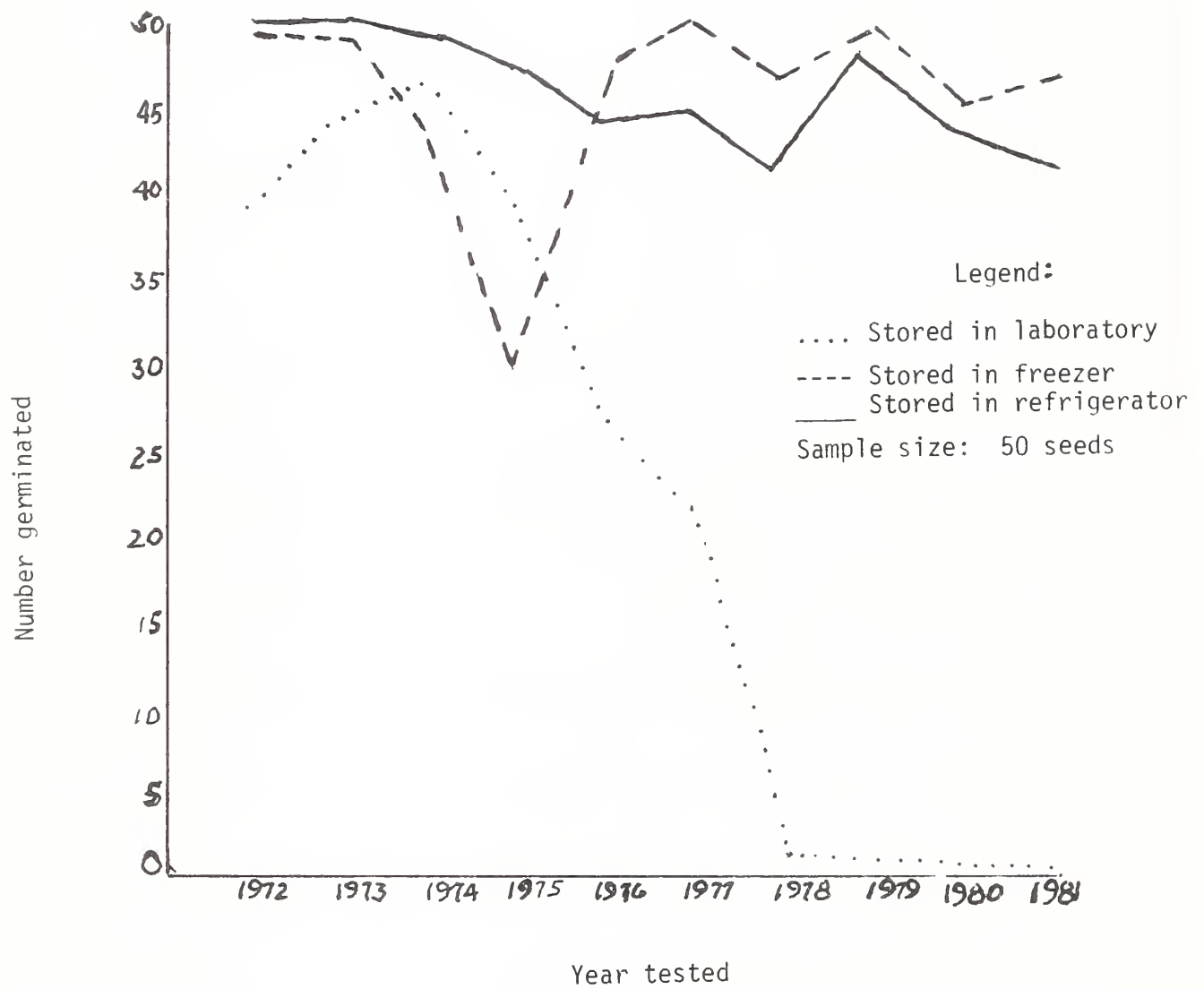


Figure 1. The effects of temperature on longevity and vitality of soybean seeds.



showed an increase in germination percentage for the years 1972 through 1974. Subsequent testings of Sample A, however, showed a consistent decrease each year until 1980 when the germination percentage reached zero. Since there was zero germination in Sample A again in 1981, a two-year period of zero germination convinced the writer to terminate the investigation. It should be noted that longevity tests of seeds in samples B and C will be continued beyond the 10-year storage period.

Based upon the results of this study, it seems reasonable to conclude that longevity is increased when soybean seeds are maintained under uniformly lower temperature conditions than those that prevail in the laboratory.

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1) Phytoestrogens in wild perennial relatives of the soybean

A systematic survey of phenolic compounds in the leaves of species from the genus *Glycine* subgenus *Glycine* Willd. (Hymowitz and Newell, 1981) is currently in progress in this laboratory. Two phenolic compounds, genistin (genistein-7-glucoside) and coumestrol, have long been known to have estrogenic activity (Bickoff et al., 1969) and both compounds have been reported from annual species of the genus *Glycine* (Wada and Masataka, 1964; Walz, 1931).

Coumestrol and genistin were isolated and identified from leaves of *Glycine tabacina* using the methods of Mabry et al. (1970). Genistin was identified by U.V. and Rf data. The sugars hydrolyzed from genistin were identified by the methods discussed by Crawford (1973). The aglycone of genistin, genistein, was confirmed chromatographically by running it on paper alongside authentic genistein (Sigma Chemical Company lot 109C-0464).

Coumestrol was identified by its U.V. and Rf data in comparison with authentic coumestrol (Eastman Kodak cat. no. 134-2799). Coumestrol from *G. tabacina* was confirmed using a Beckman HPLC and comparing its retention time at 342 n.m. with authentic coumestrol. Both compounds came off the column after 39 mins. [Altex ultrasphere O.D.S. (Reverse phase C<sub>18</sub>). Solvent 30% of 16% BuOH, 82% MeOH, 2% HoAc in 0.018M NH<sub>4</sub>OAc and 70% of 98% HoAc, 2% HoAc in 0.018M NH<sub>4</sub>OAc and flow rate 1 ml/min.] A summary of U.V. and Rf data for these compounds isolated from *G. tabacina* is given in Tables 1-4.

These compounds have also been confirmed in some other species of the genus *Glycine* subgenus *Glycine* by co-chromatography on Whatman No. 1 paper in B.A.W. and 15% HoAc (Table 5).

It is not known whether there are quantitative or qualitative differences in these compounds from species to species or within species from one region to another. Genistin does not appear to be present in *G. clandestina* (PI 440947). Coumestrol, being a bright fluorescent pink/purple color in U.V. light, could be a useful genetic marker.

There are several reports on herbarium specimens that the leaves of some of these species in Australia are eaten by animals. However, since none of these species forms large stands, they cannot be considered a significant forage and, thus, the estrogenic effect of the compounds reported here is unlikely to be of importance.

The detection of genistin and coumestrol in this subgenus of *Glycine* reinforces recent success at hybridization between subgenus *Glycine* and subgenus *Soja* (Moench) F. J. Herm. (Newell and Hymowitz, n.d.) suggesting a strong relationship between the perennial species of *Glycine* and the soybean.

Voucher herbarium specimens of the accessions mentioned in this paper are deposited in the Crop Evolution Laboratory herbarium (CEL) and seed of these accessions are maintained by the Agronomy Department, University of Illinois at Urbana-Champaign.

Table 1. R<sub>f</sub> and R<sub>rutin</sub> values of genistin from *Glycine tabacina*

Solvent	R <sub>f</sub> value <sup>a</sup>	R <sub>rutin</sub> value <sup>b</sup>
15% HoAc	0.61	1.11
B.A.W. <sup>c</sup>	0.76	1.20
B.E.W. <sup>d</sup>	0.70	1.55

<sup>a</sup>Distance moved by compound in relation to solvent front. Solvent 15% HoAc was run on Whatman No. 1 paper, B.A.W. and B.E.W. was run on Whatman No. 3 paper.

<sup>b</sup>Distance moved by compound in relation to rutin marker (Rutin from Sigma Chemical Company lot No. 109C-0464).

<sup>c</sup>B.A.W. is 4:1:2 (n-butanol; glacial acetic acid; water).

<sup>d</sup>B.E.W. is 4:1:2.2 (n-butanol; ethanol; water).

Table 2. U.V. spectral data ( $\lambda$  max. n.m.)<sup>a</sup> of genistin from *Glycine tabacina*

Methanol <sup>b</sup>	263, 352 (sh)
NaOMe	270, 366 (sh)
AlCl <sub>3</sub>	275, 305 (sh), 381
AlCl <sub>3</sub> + HCl	275, 305 (sh), 381
NaOAc	266, 324 (sh)
NaOAc + H <sub>3</sub> BO <sub>3</sub>	266, 326 (sh)

<sup>a</sup>Spectrometer Hitachi model 110.

<sup>b</sup>Different solutions in which compounds spectral data is taken.

Table 3. R<sub>f</sub> and R<sub>rutin</sub> values of coumestrol from *Glycine tabacina*

Solvent	R <sub>f</sub> value	R <sub>rutin</sub> value
15% HoAc	0.10	0.17
B.A.W.	0.82	2.45
B.E.W.	0.88	2.01

Table 4. U.V. spectral data ( $\lambda$  max. n.m.) of coumestrol from *Glycine tabacina*

Methanol	224, 250, 310, 349
NaOMe	224, 258, 320, 380
AlCl <sub>3</sub>	221, 246, 306, 344
AlCl <sub>3</sub> + HCl	219, 246, 306, 366
NaOAc	248, 266 (sh), 313, 351
NaOAc + H <sub>3</sub> BO <sub>3</sub>	246, 307

Table 5. Perennial wild species of *Glycine* in which genistin and coumestrol have been detected

	<u>Genistin</u>	<u>Coumestrol</u>
<i>Glycine tabacina</i> (PI 440991)	+	+
<i>Glycine tomentella</i> (PI 441002 x PI 373980)	+	+
<i>Glycine canescens</i> (PI 440938)	+	+
<i>Glycine clandestina</i> (PI 440947)	?	+
<i>Glycine falcata</i> (PI 440975)	+	+

<sup>+</sup> Presence confirmed.

## Acknowledgments

We would like to thank Dr. W. L. Banwart and Paul Porter for use of and help with HPLC.

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## 2) 245 Root fluorescence in the genus *Glycine* subgenus *Glycine* [ ]

Root fluorescence appears to be a useful trait for the characterization of genetic diversity in *Glycine max* and *G. soja* (Broich, 1978; Delannay and Palmer, 1982). The objective of the research reported herein was to determine the diversity of root fluorescence in the seven wild perennial species in the subgenus *Glycine* (Hymowitz and Newell, 1981).

To screen for root fluorescence, 5 or more seeds of each accession were scarified, germinated on moist filter paper in a 100 x 15 mm petri dish and placed in the dark. After several days, the roots were observed under a 365 nm long wave UV light source. The roots were classified as either fluorescent (+) or nonfluorescent (-). 'Minsoy' was used as the standard nonfluorescent type (Fehr and Giese, 1971).

Of the 172 accessions tested, nonfluorescent genotypes were found in *Glycine canescens*, *G. clandestina* and *G. tomentella* (Table 1). The accessions without root fluorescence are listed in Table 2. Within *G. clandestina*, 5 accessions had an unusual fluorescent hue. Preliminary investigations utilizing a Perkin-Elmer fluorescence spectrophotometer revealed that,



at 320 mm excitation energy, the emission peak was shifted from 420 nm in the normal fluorescent type to 440 nm in the unusual *G. clandestina* type. In addition, all of the unusual *G. clandestina* fluorescent types had curved pods and were collected from Brampton Island, Queensland, Australia.

Table 1. Accessions of wild perennial *Glycine* species with (+) or without (-) root fluorescence

Species	Accession number	Fluorescence	
		+	-
<i>G. canescens</i>	20	12	8
<i>G. clandestina</i>	42	34*	8
<i>G. falcata</i>	3	3	0
<i>G. latrobeana</i>	2	2	0
<i>G. latifolia</i>	9	9	0
<i>G. tabacina</i>	65	65	0
<i>G. tomentella</i>	<u>31</u>	<u>27</u>	<u>4</u>
Total	172	152	20

\*Unusual fluorescent genotypes: P.I. 440962, P.I. 440963, P.I. 440964, P.I. 440965 and P.I. 446944.

Table 2. Accessions without root fluorescence

Species	Accession Number
<i>G. canescens</i>	PI 399478, PI 440927, PI 440931, PI 440935, PI 440936, PI 440942, PI 446934, UI 633 (PI number not assigned)
<i>G. clandestina</i>	PI 339656, PI 440955, PI 440957, PI 440958, PI 440959, PI 440960, PI 440967, PI 446943
<i>G. tomentella</i>	PI 339657, PI 441007, PI 441009, PI 441010

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## <sup>10245</sup> Genetic analysis of a chlorophyll deficient, tan-saddle mutant [J].

In the 1961 Uniform <sup>✓</sup>[Soybean] Test II, seeds with a tan-saddle pattern were found among normally yellow-seeded 'Harosoy' plants. The tan-saddle pattern was found to breed true, and is now designated  $k_2$ . The Harosoy line from which  $k_2$  was derived is designated T239 in the Genetic Type Collection.

Among later generations of T239 some plants were found that were chlorophyll deficient. The chlorophyll deficient trait also bred true. One of the progeny of these chlorophyll deficient, tan-saddle plants was harvested and was designated T253 ( $cd-k_2$ ).

In 1965, an independent mutation to  $k_2$  and to 'chlorophyll deficient' occurred in L67-4323 (suspect  $cd-k_2$ ). Because of differing times of greening of  $cd-k_2$  and L67-4323, it has been suggested that the chlorophyll deficiency found in L67-4323 might be different from the chlorophyll deficiency found in T253 (R. L. Bernard, personal communication).

Our objective was to determine if the mutation causing the tan-saddle seeds and the chlorophyll deficiency in the suspect  $cd-k_2$  was the same as the mutations in  $k_2$  (T239) and in  $cd-k_2$  (T253).

We made reciprocal crosses with  $k_2$  and suspect  $cd-k_2$  and reciprocal crosses with  $cd-k_2$  and suspect  $cd-k_2$ . In addition, reciprocal crosses were made between the suspect  $cd-k_2$  and  $cyt-Y_2$ , a new cytoplasmic mutation affecting chlorophyll development (Palmer and Mascia, 1980). It is known that the  $cd-k_2$  mutant (T253) interacts with  $cyt-Y_2$  (Palmer and Cianzio, unpublished). The crosses with  $cyt-Y_2$  were made to determine if the suspect

$cd-k_2$  interacts in the same manner as the known  $cd-k_2$  in the presence of  $cyt-Y_2$ .

When the suspect  $cd-k_2$  was crossed as a male parent with  $k_2$  only tan-saddle seeds were found among the progeny in the  $F_1$  and in the  $F_2$  (Table 1). In addition,  $F_2$  progeny segregated 3 green : 1 chlorophyll deficient, thus confirming the hybrid origin of the  $F_2$  (Table 1). In this cross, and others, obvious 'outliers' and plots not exhibiting evidence of hybrid origin, were not included in the analysis. These results show that the mutation causing tan-saddle seeds in the suspect  $cd-k_2$  is the same as the mutation causing tan-saddle seeds in T239.

When the suspect  $cd-k_2$  was crossed as a female parent with  $k_2$ , again, only tan-saddle seeds were found in the  $F_1$  and in the  $F_2$  (Table 2).  $F_2$  progeny segregated 3 green : 1 chlorophyll deficient, which again confirmed the hybrid origin of the  $F_2$  (Table 2). In this cross, the ratio of green : chlorophyll deficient more represents a 4:1 or a 5:1 segregation than a 3:1 segregation, but this is due mainly to the effects of an early July hail-storm that struck the  $F_2$  plants of this cross before the chlorophyll deficient plants could be identified and tagged. The chlorophyll deficient plants, being weaker, were unable to survive partial defoliation and as a result many died before being identified as chlorophyll deficient.

When the suspect  $cd-k_2$  was crossed as a male parent with the known  $cd-k_2$ , all  $F_1$  and all  $F_2$  progeny were both chlorophyll deficient, and possessed tan-saddle seeds (Table 1). These results allow us to conclude that the mutations for chlorophyll deficiency and for tan-saddle seeds are the same in both L67-4323 and T253. Similar results were obtained from reciprocal crosses (Table 2). In both types of crosses,  $W_1$  (purple flower) and  $w_1$  (white flower) were used as genetic markers. Among the  $F_2$  of the reciprocal crosses, flower color segregated 3 purple : 1 white, confirming that the progeny were the result of a hybridization (Tables 1 and 2).

When the suspect  $cd-k_2$  was used as a male parent in crosses involving  $cyt-Y_2$ , all  $F_1$  progeny were yellow (Table 1). All  $F_2$  progeny were also yellow, but none of the  $F_2$  possessed tan-saddle seeds (Table 1). The absence of tan-saddle seeds in the  $F_2$  in the presence of  $cyt-Y_2$  is the same phenomenon noted in the nuclear-cytoplasmic interaction between the known  $cd-k_2$  and  $cyt-Y_2$  (Palmer and Cianzio, unpublished). In this cross, pubescence color was used as the genetic marker and the 3 dominant : 1 recessive segregation (Table 1) in the  $F_2$  population confirmed the hybrid origin of the  $F_2$ .

When the suspect  $cd-k_2$  was crossed as a female parent with  $cyt-Y_2$ , no chlorophyll deficient plants, and no tan-saddle seeds were observed in the  $F_1$  (Table 2). Among the  $F_2$ , progeny segregated 3 green, non-saddle : 1 chlorophyll deficient, tan-saddle (Table 2). Tawny pubescence and gray pubescence were used as genetic markers and segregation for these traits was also 3 dominant : 1 recessive (Table 2). If considered as a dihybrid, the segregation pattern in this cross was 9 green, non-saddle, tawny : 3 green, non-saddle, gray : 3 chlorophyll deficient, tan-saddle, tawny : 1 chlorophyll deficient, tan-saddle, gray (Table 2).

The results of our crosses have shown that the mutations responsible for the chlorophyll deficiency and the tan-saddle seeds of plants derived from L67-4323, and of plants derived from T239 and T253, are the same. It would, therefore, be inappropriate to assign a new Genetic Type Collection Number to L67-4323.

Table 1. Crosses involving the suspect  $cd-k_2$  as a male parent with soybean mutants  $k_2$ ,  $cd-k_2$ , and  $cyt-Y_2$

Cross	F <sub>1</sub>	F <sub>2</sub>
$k_2$ x suspect $cd-k_2$	all green plants	plants segregated 344 green : 95 chlorophyll deficient (3:1) $\chi^2 = 2.64$ , $P < 0.25 > 0.10$
	all tan-saddle seed	all tan-saddle seed
$cd-k_2$ x suspect $cd-k_2$	all chlorophyll deficient	all chlorophyll deficient
	all tan-saddle seed	all tan-saddle seed
		plants segregated for flower color (3:1)
$cyt-Y_2$ x suspect $cd-k_2$	all yellow plants	all yellow plants
	all non-saddle seed	all non-saddle seed
		plants segregated 805 tawny : 237 gray (3:1) $\chi^2 = 2.83$ , $P < 0.10 > 0.05$

Table 2. Crosses involving the suspect  $cd-k_2$  as a female parent with soybean mutants  $k_2$ ,  $cd-k_2$ , and  $cyt-Y_2$

Cross	F <sub>1</sub>	F <sub>2</sub>
suspect $cd-k_2$ x $k_2$	all green plants	plants segregated 186 green : 40 chlorophyll deficient (3:1) $X^2 = 6.42$ , $P < 0.025 > 0.01$
	all tan-saddle seed	all tan-saddle seed
suspect $cd-k_2$ x $cd-k_2$	all chlorophyll deficient	all chlorophyll defi- cient
	all tan-saddle seed	all tan-saddle seed  plants segregated for flower color (3:1)
suspect $cd-k_2$ x $cyt-Y_2$	all green plants	plants segregated 503 green, non-saddle : 142 chlorophyll deficient
	all non-saddle seed	tan-saddle (3:1) $X^2 = 3.06$ , $P < 0.10 > 0.05$
		plants segregated 502 tawny : 143 gray (3:1) $X^2 = 2.75$ , $P < 0.10 > 0.05$
		plants segregated 391 green, non-saddle, tawny : 112 green, non-saddle, gray : 111 chlorophyll deficient, tan-saddle, tawny : 31 chlorophyll deficient, tan-saddle, gray (9:3:3:1) $X^2 = 5.82$ , $P < 0.25 > 0.10$

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2) <sup>245</sup> A duplicate-deficient line in soybeans,

Satellite chromosomes involved in interchanges, because of their distinct morphology, are useful for special problems. Burnham (1950) determined the frequency of alternate:adjacent 1: adjacent 2 segregation in spore quartets in maize. Kunzel and Nicoloff (1979) modified the karyotype of barley (*Hordeum vulgare* L.) by inducing interchanges in order to distinguish the seven chromosomes from each other, and Langer and Kaul (1979) described an aberrant nucleolar-organizing region in *Allium cepa* L. in which the NOR consists of a fine heterochromatin stalk terminating into a deep-staining satellite.

The satellite chromosome in soybeans can be identified in root tip cells (Palmer and Heer, 1973). It has a prominent secondary constriction separating a small satellite. Although the centromeric constriction is not evident in most of the satellite chromosomes observed, a few chromosomes in which the constriction is distinct indicate that the satellite is on the short arm of the chromosome. No other mitotic chromosomes of the standard complement in soybeans have been identified.

From radiated soybeans (Sadanaga and Grindeland, 1979), three lines with altered satellite chromosomes have been developed. Line 172-11-3 in 'Hodgson' has a reciprocal translocation in which the interchanged chromosomes are identifiable. The interchanged satellite chromosome is short and the other interchanged chromosome is long.

A second line, 175-7-3, derived from 175-7 in cultivar 'Steele', has two chromosomes with a satellite that is 3 to 5 times longer than the standard satellite and two short chromosomes. These two pairs of identifiable chromosomes suggest a reciprocal exchange of asymmetrical chromosome segments. An alternative hypothesis is that a chromosome morphologically similar to that postulated from a chromosome interchange arose through an inversion in the short arm with one break in the satellite. Under this hypothesis, the short chromosomes are assumed to be either centric fragments or a pair of interchanged chromosomes that had exchanged segments with a nonsatellite chromosome.

The third line, 175-7-8, derived from 175-7 in Steele, is shorter and matures later than 175-7-3, and its flowers tend to be cleistogamous. Root tip squashes of 175-7-8 revealed two satellite chromosomes with a long satellite as in 175-7-3 but without short chromosomes.

We report the pollen and ovule sterility and chromosome associations in selected parents and hybrids to determine whether 175-7-3 is homozygous for a reciprocal translocation and 175-7-8 is a duplicate-deficient line.

Cross: 175-7-3 x Steele and reciprocal. The average pollen and ovule sterility observed in reciprocal hybrids was 26% and 45.1%, respectively (Table 1). Quadrivalents observed in metaphase I (MI) were either a ring or a chain. A small univalent chromosome observed in the PMC's with a trivalent was the short chromosome. In some PMC's at anaphase I (AI), the short chromosome lagged at the equatorial plate. No anaphase bridges or fragments were observed. The quadrivalent observed in the PMC's and pollen sterility indicated that 175-7-3 is homozygous for a reciprocal translocation.

Cross: 175-7-3 x 175-7-8. The frequency of the different kinds of chromosome associations is shown in Table 1. There was a higher frequency of bivalents and a lower percentage of sterile pollen than in the previous cross. The univalent observed in the PMC was a short chromosome.

Cross: 175-7-8 x Steele. The average pollen sterility in three hybrids of this cross were not significantly different from the pollen sterility in progeny of selfed 175-7-8 (Table 1). Fertile pollen are expected from these hybrids regardless of bivalent or quadrivalent association. Parental chromosomes are expected from alternate and adjacent-1 disjunctions in the quadrivalent.

Origin of 175-7-8. In the translocation heterozygote of 175-7-3, three of the four chromosomes involved in an association of four can be identified in root tip mitotic cells. These are the chromosome with the large satellite, the chromosome with the standard satellite, and the short chromosome. In line 175-7-8, we observed two nucleolar chromosomes with a large satellite but no short chromosomes. Plants of this chromosome constitution can arise from the union of two duplicate-deficient gametes carrying the chromosome with the large satellite (interchanged chromosome) and the standard nonsatellite chromosome involved in the interchange. Cytological analysis of root tip cells of  $F_2$  progeny of a cross between 175-7-3 and T93A indicate that duplicate-deficient gametes are transmitted either through the egg or pollen (unpublished). We conclude that 175-7-8 is a duplicate-deficient line. It is tetrasomic for the interchanged segment on the nucleolar chromosome and deficient for part or all of the small satellite in the standard nucleolar chromosome.

In a cross, 175-7-8 ( $Y_7Y_8$ ) x T138 ( $y_7y_8$ ), the  $F_2$  ratio of 90 green: 6 yellow fit the expected 15:1 and indicated that neither the  $y_7$  nor  $y_8$  locus is on the interchanged segment.

Table 1. Chromosome association, pollen and ovule sterility in hybrids involving 175-7-3 and 175-7-8

Identity	Chromosome association			Sterility %	
	20 <sup>II</sup>	18 <sup>II</sup> +1 <sup>III</sup> +1 <sup>I</sup>	18 <sup>II</sup> +1 <sup>IV</sup>	pollen	ovule
175-7-3 x Steele	11	3	28	26.0	45.1
175-7-3 x 175-7-8	24	1	23	21.8	
175-7-8 x Steele	-- <sup>a</sup>	--	--	5.7	2.0
175-7-8 x T138	--	--	--	3.3	8.5
175-7-8 selfs	--	--	--	7.7	

<sup>a</sup>Not analyzed.

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X. Delannay

### 3) <sup>245</sup> A dwarf mutation in 'Hodgson' soybean.

A dwarf mutation was found in a line derived from radiated 'Hodgson' grown at the Bruner Farm near Ames, IA. The mutant plants were 6 to 10 cm tall, had necrotic leaves, and produced no seeds.

Twenty-four plants randomly picked from segregating plots were progeny-tested in a greenhouse and in the field. The field test data are shown in Table 1. The ratio of normal: dwarf plants in all segregating progeny rows except one fit a 3 normal: 1 dwarf expected for a simple recessive trait. Progeny rows that segregated dwarfs also segregated fertile and semisterile plants. Subsequent testing showed that fertile plants produced only fertile progeny and semisterile plants segregated dwarf, fertile, and semisterile progeny. Dwarfness was linked to semisterility.

Dwarf plants grown in a greenhouse grew 15 to 20 cm tall. We noticed among progeny of semisterile plants, six seedlings with light yellow unifoliate leaves. These chlorophyll-deficient seedlings subsequently developed into dwarf plants. Evidence of necrosis first appeared along the margins of the unfolding trifoliate leaves. The trifoliate leaves of branches were lanceolate and much reduced in size. Almost all floral buds on the dwarf plants were abnormal. However, a few buds bloomed, and about a dozen seeds were harvested from the six dwarf plants. Seeds from the dwarf plants produced dwarf progeny.

The gene for dwarfness was located to the interchange chromosome. F<sub>2</sub> populations of hybrids between 'Hark' and semisterile plants were of two kinds, those that produced all fertile progeny and those that segregated dwarf, semisterile, and fertile plants (Table 2). Dwarf plants, therefore, are homozygous recessive for the dwarf gene and homozygous for the translocation. Hybrids between fertile plants and Hark were all fertile.

The dwarf and chlorophyll-deficiency traits may be controlled by two genes very tightly linked or may be due to pleiotropy of one or the other. No crossover types have been observed in segregating populations, and it has not been determined whether tightly linked genes or pleiotropy control dwarfness and chlorophyll deficiency.

Forty- and 41-chromosome plants were found in progeny of a semisterile plant. In the 40-chromosome group, 18 plants were normal, 9 were dwarfs, and in the 41-chromosome group, 6 plants were normal and 0 plants were dwarf. Interchange chromosomes were not identified in root tip cells. A quadrivalent observed in pollen mother cells of a semisterile plant supported the genetic evidence of the presence of a reciprocal translocation.

Table 1. Segregation of normal and dwarf plants in 13 of 24 randomly picked plants

Identity <sup>+</sup>	Normal	Dwarf	Total	X <sup>2</sup>	Probability
35-1	99	32	131	0.02	.50 - .70
35-3	128	25	153	6.12	.01 - .02
35-5	66	26	92	0.52	.30 - .50
35-8	147	46	193	0.14	.70 - .80
36-1	123	31	154	1.95	.10 - .20
36-2	112	32	144	0.59	.30 - .50
36-4	126	43	169	0.02	.80 - .90
36-7	120	31	151	1.61	.20 - .30
39-3	99	26	125	1.18	.20 - .30
39-4	62	20	82	0.02	.50 - .70
39-6	30	11	41	0.07	.70 - .80
39-7	47	16	63	0.01	.90 - .95
39-8	<u>91</u>	<u>27</u>	<u>118</u>	<u>0.28</u>	.50 - .70
	1250	366	1616	12.53	

Chi-square

Total	12.53	
Deviation	4.76	.02 - .05
Heterogeneity, df = 12	7.77	.80 - .90

<sup>+</sup>11 plants were homozygous normal.

Table 2.  $F_2$  segregation in crosses of semisterile and fertile plants x Hark

Identity	Fertility	Normal	Dwarf	$\chi^2$	P
R47-2-1	semisterile	27	10	0.08	.30 - .50
R47-2-1 x Hark -1		175	0		
" x " -2		180	0		
R47-3	semisterile	36	7	1.74	.10 - .20
R47-3 x Hark		147	44	0.39	.50 - .70
R47-3-1	fertile	40	0		
R47-3-1 x Hark -1		112	0		
" x " -2		173	0		
" x " -3		183	0		
R47-11	semisterile	25	8	0.01	.90 - .95
R47-11 x Hark		212	52	3.96	< .05
R47-15	semisterile	32	8	0.53	.30 - .50
R47-15 x Hark -1		90	0		
" x " -2		51	0		

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#### 4) Chlorophyll-deficient plants in a soybean cross,

In 1977, 18 yellow plants were found in an  $F_2$  population of a cross between two strains of soybeans, T235 x PI 86024, both with normal green foliage. Segregation fit a ratio of 15 green: 1 yellow (Table 1). All  $F_3$  seedlings grown from seeds harvested from yellow  $F_2$  plants turned yellow as the plants grew. Yellowing, beginning about 4 to 5 weeks after germination, proceeded from the older to the younger leaves.

The phenotype of the  $F_2$  yellow plants and their  $F_3$  progeny was similar to that of strains homozygous for  $g$  and  $y_3$ , a genotype characterized by yellowing as the plant grew (Bernard and Weiss, 1973). Appropriate crosses were made to test whether the yellow segregates carried  $y_3$ . This note presents the results of the tests to determine the alleles in the yellow  $F_2$  progenies and in the parents from which they originated and linkage tests of  $y_3$  with four translocation lines.

The parents homozygous for  $G$  and  $y_3$  are PI 86024 and 'Kura', for  $g$  and  $y_3$  are T139, selection 7628 in the original cross, and L63-2346, and for  $g$  and  $Y_3$  are T235, 'Kent', L61-4222, L61-4558, 171-31-2, 172-11-3, Clark T/T, and L75-0283-4. Lines 171-31-2 and 172-11-3 are homozygous translocations found in radiated 'Hodgson' (Sadanaga and Grindeland, 1979); 'Clark T/T', developed by R. G. Palmer, is near-isogenic Clark incorporating a translocation from PI 101404B (*G. soja*); and L75-0283-4 is a spontaneous translocation found by R. G. Palmer in an  $F_4$  progeny row of a 'Beeson' x 'Amsoy 71' cross from Illinois in 1975.

All  $F_2$  plants of crosses with the translocations were grown in the greenhouse except for crosses with 172-11-3. Semisterile (translocation heterozygote)  $F_2$  plants grown in the greenhouse were identified by staining pollen grains with  $I_2KI$ . Field-grown plants were classified semisterile or fertile on the basis of number of pods and seeds per pod.

Chi-squares to test linkage between  $y_3$  and the breakpoints in the translocations were calculated according to the method of Kramer (1954).

**Results and Discussion.** The hypothesized segregation ratios in the  $F_2$  generation and the associated chi-square probabilities of the different crosses are shown in Table 1. The cross between T235 x PI 86024 again yielded a ratio of 15 green : 1 yellow seedlings. All yellow plants, without exception, were yellow seeded. The absence of yellow plants with green seed coat suggested that the  $F_2$  ratio was not due to duplicate factors. The cross L61-4222 x PI 86024 gave an  $F_2$  ratio of 15 green : 1 yellow; the cross T235 x Kent gave all green  $F_2$  plants. These results indicated that T235 carried the same alleles as L61-4222 and Kent, whereas PI 86024 carried contrasting alleles.

PI 86024 resembles Kura, a cultivar in which the inheritance of seed coat color and chlorophyll deficiency is known. Terao and Nakatomi (1929) first reported the effects of the genes  $H h$  and  $C c$ , now symbolized as  $G g$  and  $Y_3 y_3$ .  $G$  is epistatic to  $y_3$  so that hybrids between Kura and yellow-seeded green plants yield 15 green : 1 yellow seedling. Bernard and Weiss (1973) noted, "Several green-seeded Japanese varieties have the  $G y_3$  genotype, e.g., 'Kurakake' (Kura or PI 243526 in the USDA soybean collection). Therefore, 1/16 chlorophyll-deficient  $F_2$  plants ( $g y_3$ ) are often observed in breeding populations involving one parent with green seed coat." That PI 86024 may carry  $G$  and  $y_3$  was surmised from its resemblance to Kura and that both had been introduced from the same region in Japan. If PI 86024

carries the same alleles as Kura, one expects no  $F_2$  segregation for chlorophyll deficiency. The absence of segregating  $F_2$  progeny in the cross Kura x PI 86024 (Table 1) supported the hypothesis that  $G$  and  $y_3$  are in PI 86024 and that the genotype of the yellow  $F_2$  seedlings from the cross T235 x PI 86024 is  $g g y_3 y_3$ .

In the crosses 7628 x L61-5448 (Table 1) and 7628 x 172-11-3, 7628 x L75-0283-4 and reciprocal (Table 2), segregation was observed for foliage color but not for seed coat color. Selection 7628, therefore, carries the  $y_3$  allele. In cross 7628 x Kura (Table 1), on the other hand, segregation was observed for seed coat and foliage color. All green  $F_2$  plants had green seed coat and all yellow plants had yellow seed coat. In the  $F_3$  generation, 2/3 of the green plants segregated for seed coat and foliage color and 1/3 were homozygous green for seed coat and foliage color. Yellow  $F_2$  seedlings bred true, always producing seeds with yellow seed coat. In crosses between 7628 x T139 and L63-2346, yellow  $F_1$  hybrids with traits characteristic of the  $g g y_3 y_3$  genotype confirmed that 7628 has  $g$  and  $y_3$ .

The  $y_3$  locus is not listed on any of the eight linkage groups (LG) reported by Stelly and Palmer (1977);  $G$  is on LG3. Nonsignificant chi-square values indicated  $y_3$  was not linked to either of the interchanged chromosomes in translocation lines 171-31-2, 172-11-3, Clark T/T, and L75-0283-4 (Table 2). Cytological observations in translocation x translocation crosses (unpublished) indicated that translocation lines 171-31-2 and L75-0283-4 have one common chromosome involved in the interchange; translocation lines 172-11-3 and Clark T/T, also, have one common chromosome involved in the interchange. The  $y_3$  locus, therefore, was tested for linkage to six different chromosomes involved in the interchanges in the four translocation lines. The only known linkage is  $ms_1$  on LG 8 to the breakpoint in Clark T/T (Palmer, 1976). White flower color ( $w_1$ ), also on LG 8, was independent of the breakpoint. Recently, Hildebrand et al. (1980) reported that LG 9 has genes controlling two chemical components,  $Ap$  for acid phosphatase and  $Ti$  for Kunitz trypsin inhibitor, linked with a crossover frequency of 16.2%. PI 86024, which carries the  $Ti^b$  allele (Orf and Hymowitz, 1977, 1978) and other mutant genes, may be useful in linkage studies.

Table 1. Segregation of green and yellow plants in  $F_2$  populations of crosses between green x green, yellow x green, and yellow x yellow parents

Cross	Green	Yellow	Chi-square probability	
			15:1	3:1
<u>Green x Green</u>				
T235 x PI 86024	384	18	.20 - .10	
L61-4222 x PI 86024	118	6	.70 - .50	
T235 x Kent	604	0	.000	
Kura x PI 86024	88	0	.02 - .02	

Table 1. *Continued*

Cross	Green	Yellow	Chi-square probability	
			15:1	3:1
<u>Yellow x Green</u>				
7628 x L61-5448	188	50		.20 - .10
7628 x Kura	165	52		.80 - .70
<u>Yellow x Yellow</u>				
T139 x 7628	0	346		
7628 x L63-2346	0	323		

Table 2. Observed  $F_2$  segregation of  $y_3$  and the breakpoint in four translocations and their linkage chi-square probability

Cross	Semisterile		Fertile		Chi-square
	Green	Yellow	Green	Yellow	P
7628 x 172-11-3	82	33	84	23	.30 - .20
172-11-3 x T139	<u>99</u>	<u>24</u>	<u>91</u>	<u>26</u>	<u>.70 - .50</u>
Total	181	57	175	49	.70 - .50
7628 x L75-0283-4	47	11	44	13	.70 - .50
L75-0283-4 x 7628	<u>31</u>	<u>10</u>	<u>37</u>	<u>11</u>	<u>.90 - .80</u>
Total	78	21	81	24	.90 - .80
T139 x Clark T/T	62	19	72	17	.50 - .30
L63-2346 x 171-31-2	112	35	95	39	.50 - .30

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245  
3) Identifying translocations in soybeans

Six translocations, currently used in linkage studies of marker genes on known linkage groups, have been intercrossed and are being examined cytologically to identify them. The origin of these six translocations are shown in Table 1.

The identification of the translocations is based on the chromosome association of the interchange chromosomes. Two quadrivalents, a quadrivalent and a trivalent + univalent, or two trivalents + two univalents would be expected in the PMCs of  $F_1$  hybrids if the two translocations are different. A ring or chain of six chromosomes would be expected in the PMCs of  $F_1$  hybrids if the two translocations have one common chromosome involved in an interchange.

The percent of sterile pollen and chromosome associations in T x T crosses are shown in Table 2.

The translocation in Clark T/T is different from that in L75-0283-4, PI 189866, and 171-31-2. One common chromosome is involved in an interchange in Clark T/T, 172-11-3, and 175-7-3.

The translocation in L75-0283-4 is different from that in PI 189866, 172-11-3, and 175-7-3. One common chromosome is involved in an interchange in L75-0283-4 and 171-31-2.

PI 189866 and 172-11-3, 172-11-3 and 171-31-2, and 171-31-2 and 175-7-3 have different translocations. One common chromosome (satellite chromosome) is involved in an interchange in 172-11-3 and 175-7-3.

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Table 1. Origin of six translocations in soybeans

Translocation	Origin
Clark T/T	Near-isogenic Clark with translocation from PI 101404B incorporated. PI 101404B is introduction from NE China.
L75-0283-4	Spontaneous translocation in an F <sub>4</sub> progeny row of a Beeson x Amsoy 71 cross. Found by R. G. Palmer in Illinois in 1975.
PI 189866	<i>Glycine gracilis</i> introduction from NE China.
171-31-2	Translocation from a radiated population of Hodgson. Selected by K. Sadanaga.
172-11-3	Translocation from a radiated population of Hodgson. Selected by K. Sadanaga.
175-7-3	Translocation from a radiated population of Steele. Selected by K. Sadanaga.

Table 2. Percentage of aborted pollen and chromosome associations in T x T crosses

Cross	Aborted pollen (%)			Chromosome association
	KN 1979 <sup>a</sup>	KS 1980	KS 1981	
Clark T/T X				
L75-0283-4	74.0 ± 5.5	66.6 ± 2.0		2 IV
PI 189866	78.3 ± 4.6	67.4 ± 2.4		2 IV
171-31-2		73.2 ± 2.2	75.4 ± 4.8	2 IV
175-7-3		61.2 ± 3.2	62.7 ± 4.4	1 VI
172-11-3	68.7 ± 7.0	64.9 ± 1.8	62.9 ± 1.1	1 VI
L75-0283-4 X				
PI 189866	77.9 ± 3.3	72.3 ± 2.4	72.1 ± 4.5	2 IV
175-7-3		63.9 ± 2.8	66.7 ± 3.8	2 IV
172-11-3	77.6 ± 4.7	70.1 ± 2.6	72.6 ± 5.2	2 IV
171-31-2		65.2 ± 1.3	61.7	1 VI
PI 189866 X				
172-11-3	73.5 ± 3.2	68.3 ± 3.1	70.0 ± 3.6	2 IV
175-7-3			58.8 ± 5.1	? <sup>b</sup>
171-31-2			66.1 ± 4.8	? <sup>b</sup>
172-11-3 X				
175-7-3		50.2 ± 8.5	50.9 ± 3.4	1 VI
171-31-2		74.2 ± 7.3		2 IV
171-31-2 X				
175-7-3		72.3 ± 6.0	70.6 ± 3.7	2 IV

<sup>a</sup>Pollen sterility of plants grown in greenhouse. Data of K. Newhouse.

<sup>b</sup>Analysis not complete.



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## Genetic linkage analysis <sup>47</sup><sub>15</sub> <sup>5</sup><sub>123</sub>

The  $Rj_2$  gene (Caldwell, 1966), which conditions ineffective nodulation with strains of the cl and l22 serogroup of *Rhizobium japonicum* (Kirchner) Buhanan, and the  $Rj_4$  gene (Vest and Caldwell, 1972), conditioning an ineffective nodulation response with rhizobial strain 61 of the Beltsville Culture Collection *R. japonicum*, were tested for linkage association with the gene  $L_1$  (black pod). The  $rj_1$  gene (Williams and Lynch, 1954), conditioning a non-nodulating response with a broad spectrum of strains of *R. japonicum*, was tested for linkage with the gene  $fr$  (roots nonfluorescent in UV light) and  $L_1$  (black pod). The gene  $y_9$  (chlorophyll defective) was tested for possible linkage with the genes  $ln$  (narrow leaf), and  $P_1$  (glabrous). The  $P_1$  gene was also tested for linkage with the gene  $fr$ .

Materials and Methods. Genetic stocks (T lines) and Clark  $rj_1 rj_1$  were obtained from the Soybeans Genetic Type Collection (Bernard and Weiss, 1973). Crosses were made in the field (Table 1) and  $F_1$  seed were advanced to the  $F_2$  generation in the greenhouse. In some crosses, the  $F_2$  progeny were assayed directly for expression of the pertinent phenotype. In other crosses, the  $F_3$  progeny were assayed and the  $F_2$  genotypes rationalized from the  $F_3$  phenotypes.  $F_3$  seed was produced in the field at Beltsville. Crosses with the  $Rj_2$  and  $Rj_4$  genes were evaluated in plastic growth tray assemblies (Devine and Reisinger, 1978) and inoculated with 7-day-old broth cultures of strain 7 of *R. japonicum* from the Beltsville Culture Collection to define for  $Rj_2$  and strain 61 to define for  $Rj_4$ .

The progeny of crosses T135 x T109 and T135 x T145 were evaluated for chlorosis, narrow leaf and glabrous traits in the  $F_2$  generation in the field at Beltsville. In the case of the T215 x 'Clark'  $rj_1$  cross, each  $F_3$  seed lot was divided between two packets and two 10-foot-row plantings were made in the field. The first planting was dug and the  $F_3$  plants were scored for nodulation response to determine the presence of the  $rj_1$  gene. The field had been used for soybean cultivation for many years previously and contained abundant *R. japonicum*. The second planting was allowed to mature and the progeny rows were scored for segregation of the black pod trait.

In the cross T145 x 'Minsoy',  $F_2$  seed were germinated in petri dishes and examined, 3 days after the beginning of water imbibition, under UV light for fluorescence of the radical. After classification for fluorescence, the seedlings were transplanted into rows in growth trays, cultured in a growth room, and classified for the glabrous character at the first trifoliolate-leaf stage. For the cross of Clark  $rj_1$  x Minsoy,  $F_2$  seedlings were classified for fluorescence as previously described, then were inoculated with strain 7 of *R. japonicum* and transplanted into rows in vermiculite-filled growth trays. After an additional 3 weeks growth, the seedlings were removed from the vermiculite and classified for nodulation response.

In the cross T215 x 'Hardee', each  $F_3$  seed lot was divided and about 50 seed were evaluated for the presence of the nodulation response gene  $Rj_2$  under conditions of controlled inoculation with rhizobial strain 7 in growth trays in the greenhouse. The remaining seed was planted in 10-foot rows in

the field at Beltsville, MD, and grown to maturity when rows were classified for segregation for the black pod trait. Similarly, in the 'Hill' x T215 cross, the  $F_3$  seed lots were divided and one portion of seed was characterized for the black pod trait in the field and the other portion was characterized for the  $Rj_4$  nodulation response gene in growth trays after inoculation with rhizobial strain 61.

Results. Results of the seven linkage tests are given in Table 2. All of the traits tested displayed independent assortment indicating they were not genetically linked to the genes tested.

Table 1.

Cross	Generation of progeny evaluated	Trait characterized		
		In field	In growth trays	In petri dishes
T135 ( $y_9 LN$ ) x T109 ( $Y_9 ln$ )	$F_2$	$y_9 ln$	--	--
T135 ( $y_9 p_1$ ) x T124 ( $Y_9 P_1$ )	$F_2$	$y_9 p_1$	--	--
T215 ( $L_1 Rj_1$ ) x Clark $rj_1$ ( $l_1 rj_1$ )	$F_3$	$l_1 rj_1$	--	--
T145 ( $Fr P_1$ ) x Minsoy ( $fr p_1$ )	$F_2$	--	$p_1$	$fr$
Clark $rj_1$ ( $rj_1 Fr$ ) x Minsoy ( $Rj_1 fr$ )	$F_2$	--	$rj_1$	$fr$
T215 ( $L_1 rj_2$ ) x Hardee ( $l_1 Rj_2$ )	$F_3$	$L_1$	$rj_2$	--
Hill ( $Rj_4 l_1$ ) x T215 ( $rj_4 L_1$ )	$F_3$	$L_1$	$rj_4$	--

Table 2. Soybean genetic linkage tests

Genes	a	b	c	d	Sum	%R*	SE	Phase
$Y_9 y_9 L_n l_n$	T135 ( $y_9 y_9 L_n l_n$ ) x T109 ( $Y_9 Y_9 l_n l_n$ )							
	171	45	52	14	282	50	4	R
$L_1 l_1 Rj_2 rj_2$	T215 ( $L_1 L_1 rj_2 rj_2$ ) x Hardee ( $l_1 l_1 Rj_2 Rj_2$ )							
	55	11	20	6	92	55	7	R
$Rj_4 rj_4 L_1 l_1$	H111 ( $Rj_4 Rj_4 l_1 l_1$ ) x T215 ( $rj_4 rj_4 L_1 L_1$ )							
	132	34	27	5	198	45	6	R
$Y_9 y_9 P_1 p_1$	T135 ( $y_9 y_9 P_1 p_1$ ) x T145 ( $Y_9 Y_9 P_1 P_1$ )							
	144	40	55	17	256	49	5	C
$P_1 p_1 Fr fr$	T145 ( $P_1 P_1 Fr Fr$ ) x Minsoy ( $p_1 p_1 fr fr$ )							
	291	91	102	32	516	50	3	C
$Rj_1 rj_1 Fr fr$	Clark $rj_1 (rj_1 rj_1 Fr Fr)$ x Minsoy ( $Rj_1 Rj_1 fr fr$ )							
	76	31	24	10	141	50	6	R
$L_1 l_1 Rj_1 rj_1$	T215 ( $L_1 L_1 Rj_1 Rj_1$ ) x Clark $rj_1 (l_1 l_1 rj_1 rj_1)$							
	115	31	44	14	204	48	5	R

\*Recombination percentages calculated by the product method (Immer and Henderson, 1943).

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245  
X) The genetic basis of physiological races of phytophthora [12],

All characters of living organisms, whether morphological, physiological, or biochemical, undoubtedly are genetically determined. The ability of a few microorganisms to attack a limited range of plant species and the within-species variation in disease development also are presumed to have genetic bases. Such a variation in disease expression was used to develop the gene-for-gene concept (Flor, 1955). Its development and most of its subsequent application has been with the qualitative infection types (IT's) of rusts and mildews. Ellingboe (1976) has extensively used and written about the gene-for-gene concept. Loegering (1978) used it also to develop interorganismal genetics -- the study of the interactions among the genes of the symbionts in symbiotic associations.

✓ Phytophthora [root rot] does not have distinct IT's ranging from hypersensitive 0 fleck through large IT4 pustules. Soybean hypocotyl inoculation with phytophthora mycelia or zoospores is not natural but repeatable. Plants are generally dead or healthy after several days, although there is some variation (Ward et al., 1979). However, the same basic principles of interorganismal genetics apply even though the disease phenotypes are quite different.

✓ The historical development of the *Phytophthora* [soybean association] is very similar to the cereal rusts although more recent. Laviolette and Athow (1981) summarized the history. The disease was first observed in 1948, but it was 1959 before the causal organism was named *Phytophthora megasperma* f. sp. *sojae* (now *glycinea*). Resistance of soybeans to it was identified, and it appeared to be due to a single dominant gene -- amenable for backcrossing into all available cultivars. It was no great surprise when race 2 was reported in 1965; some isolate differences in virulence had been observed previously. Thus, we had the basic pattern of one gene pair in each organism -- the corresponding gene pairs (CGP) which were the basis used to develop the gene-for-gene hypothesis.

Race		
	1	2
'Sanga'	-	+
'Harosoy'	+	+

This isn't the pattern most of you were looking for -- it will be discussed later. Ellingboe (1976) used - for incompatible and + for compatible relationships. The differences could be large or small, but detectable.

Race 3 was reported in 1972, so now we definitely had specificity superimposed on the basic compatibility necessary for the disease to develop in the first place (Ellingboe, 1976). Specificity is due to different CGPs;



	Race 2	
	2	3
'Mukden'	-	+
Sanga	+	-

the genetic basis of basic compatibility is not well-defined (Heath, 1981). Keeling (1980) reported races 10 to 16. Apparently, more are in captivity awaiting naming. There probably are at least 10 *Rps* genes. The corresponding variation in *Phytophthora* gives us  $2^{10}$ , or 1024, possible races to identify, a horrifying possibility.

Loegering (1978) objected to the use of + and - because people tend to interpret them as host susceptibility and resistance. These are useful anthropocentric terms, but detract from the pure genetics of symbioses. Loegering used Boolean algebra to eliminate any good-bad connotations of the phenotypes.

	1	0
1	1	0
0	0	0

The 1 and 0's outside the box represent the definitive (D or 1) and non-definitive (N or 0) genotypes of the two symbionts; dominance, homozygosity or ploidy are not implied. Inside the box are the phenotypes of the associations which are determined by the genotypes of both organisms. The D or 1 phenotype results only from D or 1 genotypes of both organisms, which can be represented as p (pathogen)/h (host) lp/lh = 1 phenotype. This lp/lh is the genotype of the association. An N or 0 phenotype can result from three different association genotypes, i.e., lp/Oh, Op/lh, or Op/Oh. These relationships are the basis of interorganismal genetics, and I hope to show how it can be useful with *Phytophthora*. The D or 1 phenotype is generally the desirable one (host resistant) in most associations or diseases, including phytophthora rot. Victoria blight of oats usually is given as the exception, with D = undesirable (host susceptible). Ellingboe (1976) suggests this is the evolution of the basic compatibility which genetically seems reasonable.

The pattern observed with Victoria blight of oats also can be found with phytophthora rot, although the genetics are completely different. We know that Mukden has  $Rps_1^a$  and Sanga has a non-a allele, a host gene pair

	Race	
	1	2
Mukden	-	-
Sanga	-	+

for reaction. Obviously, there is a pathogen gene pair for pathogenicity on Sanga which appears to satisfy the definition of CGP. However, the usual  $\begin{smallmatrix} -+ \\ ++ \end{smallmatrix}$  pattern and the Victoria blight exception  $\begin{smallmatrix} +- \\ -- \end{smallmatrix}$  occur with ONLY one

corresponding D genotype in each organism. Intraorganismal genetics identifies the genes which can be postulated from interorganismal D phenotypes. Distinctions sometimes appear to be arbitrary and unnecessary. We know that Sanga has  $Rps_1^b$ , a D genotype, designated here as lhlb. This means D, host, locus 1, and allele b. Similarly, Mukden is lhla. Race 1 has both corresponding definitive genotypes, lpla and lplb, which gives the association genotypes lpla/lhla and lplb/lhlb and the desirable D or l phenotype of healthy plants of Mukden and Sanga. The same association genotype is effective with race 2-Mukden. Race 2-Sanga is

$$\frac{lpla, 0plb}{lhlb}$$

and the undesirable N or 0 phenotype because the plant is dead from hypocotyl inoculation. In the field, plant injury would vary with environmental conditions. Obviously, a homozygous diploid plant cannot have 2 different alleles at a locus. Such allelism is common in the host but rare or nonexistent in the fungi; *Phytophthora* does not appear to be an exception.

The interactions might be easier to visualize if it had been demonstrated that when resistance and avirulence were dominant, induced host lectins (L) bound specific pathogen carbohydrates (C) to stop their growth. L-C binding is similar to antibody-antigen formation (Lis and Sharon, 1973) which are specific with much genetic variation. Then the Mukden Lla could interact with the complementary gene product Cla to stop development of cultures representing races 1 and 2. The Sanga Llb would bind the race 1 Clb which race 2 does not have. In the host heterozygote, neither allele "dominates" the other, in agreement with the results published. On the other hand, several other processes may be responsible for all the results.

A number of *Rps* genes have been identified. Using the definitive relationship of the association genotype (pathogen/host, p and h omitted for brevity) 1/1 = l phenotype, we can assign genotypes to *Phytophthora* from a table of single-gene host lines-races like the one Alan Walker distributed at our St. Louis meeting (1982). Race 1 has the genotype: 1la, 1lb, 1lc, 1lk, 12, 13, 14, 15, 16; race 2 differs only as 0lb and race 3 as 0la. People at Purdue are developing three-gene lines. One of these lines with race 7 would have

$$\frac{0la, 1lb, 1lc, 1k, 02, 03, 04, 05, 06}{1lb \quad \quad \quad 02, 13, 04, 05, 16}$$

with the definitive phenotype (host resistant) due the corresponding 1lb's. A mutation in *Phytophthora* to 0lb would make it capable of damaging this line with three genes for resistance. The D phenotype in this example is "epistatic" (Loegering, 1978) to all the possible N phenotypes. This also occurs where several IT's are indicated in the association genotype; genes functioning later cannot be observed when disease development ceases earlier.

$Rps_1^a$  is rather cumbersome and could be more simply written as Prla, indicating phytophthora rot locus 1, allele a. This follows the usage with leaf and stem rusts (Lr, Sr) etc., where dominance also is not indicated. Prla ( $Rps_1^a$ ) is dominant to Prlb ( $Rps_1^b$ ) with race 1, but Prlb is dominant to Prla with race 2?! These are the definitive genotypes (1la and 1lb) interacting with the corresponding pathogen 1la and 1lb. If we can specify genes, do we still need races?

Green (1965) proposed a method by which races were described by "effective/ineffective host genes" with different combinations given code numbers. Loegering and Browder (1971) changed this to a pathogen orientation with avirulence/virulence formulae, retaining the use of codes. They assigned sequential numbers to the differentials, which were not an absolute set. These numbers were to be used in the formulae. Browder et al. (1980) suggested the pathogenicity formula avirulence/virulence without any codes or numbers. They effectively argued that codes serve the same function as race designations and both should be eliminated. Using differentials with single genes does have some advantages, although sometimes additional genes are identified in putative single-gene lines. The results could be expressed with the orientation to the host (effective/ineffective), pathogen (avirulent/virulent), or both, by the association phenotype D/N. The basic definitive (D) phenotype has some advantages since it specifies the genotypes of both organisms. The two categories in all three systems are mutually exclusive, thus only one need be listed, e.g., D, when the host lines-genes are included. If all isolates from a field on six single-gene lines, e.g., Mukden (prla), Sanga (prlb) 'Arksoy' (prlc), 'Kingwa' (prlk), PI 86972-1 (Pr3) and 'Altona' (Pr6), have the D formula 1a, 1k, 3, 6, then the *Phytophthora* population has the active 1a, 1k, 3 and 6 alleles and cultivars with any one of the corresponding Pr genes would not be damaged.

The specific host genes to include in the differentials would depend on the objectives of the survey or experiment. The number of Pr genes identified is increasing, and it may be unmanageable to use them all separately. Multigene lines would help identify additional pathogen genes, but would give incomplete information about the presence of known pathogen genes due to "epistasis."

Considerable effort is necessary to determine genetic uniformity within fields and factors effecting changes. However, this must be done where Pr is a severe problem. Athow (1973) indicated that Pr did not become a problem until very susceptible cultivars were widely grown. Field or general resistance may be adequate on many soils -- didn't Hartwig use natural selection to increase it? Farmers must also consider other diseases, nematodes, lodging, yield, etc. Establishing the importance of any pest objectively is rather difficult but should be done, with the alternatives evaluated, before specific corrective measures are adopted.

We speak of specific genes (genotypes) in soybeans. A definitive association phenotype results from only the definitive genotypes of both organisms. Thus, we can identify specific *Phytophthora* genes. It seems that the genetic composition of *Phytophthora* populations should tell us more about them than arbitrary race classification. Physiological specialization may be due to a deletion or change at only one locus.

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1) <sup>145</sup> Preliminary electrophoretic observations from several soybean enzymes [F-3],

We have been studying a number of enzyme systems electrophoretically in both cultivated (*G. max*) and wild soybeans (*G. soja*), as well as in related species (Gorman and Kiang, 1977; 1978; Kiang, 1981; Gorman et al., n.d.). Our primary concern has been to measure genetic variation within and between wild and cultivated soybeans. We would like to report some miscellaneous observations from our work that have not been previously reported.

1213 A) Leucine amino peptidase (LAP): Two anodal electrophoretic bands have been observed in all *Glycine* species that we have stained for LAP. In both [*G. max*] and [*G. soja*] samples the first band, from the origin, seems to be stored in mature seeds but loses activity a couple of days after germination, while the second band is primarily seen in green tissues. The first band has been observed to have two electrophoretic forms in both *G. max* and *G. soja* accessions, one with an  $R_f$  value of .49 and the other with an  $R_f$  of .44 inversely relative to methyl blue. A single locus with two codominant alleles seems to be responsible for the difference (Table 1). No polymorphic forms have yet been observed for the second band in *G. max* or *G. soja*, but different mobility forms have been observed in *G. tomentella*. Since mobility variants of either band had no effect on the other band's mobility, and since these bands showed clear differences in tissue distribution patterns, we believe that each band was the product of unique loci.

B) Glutamate oxaloacetic transaminase (GOT): Three anodal GOT bands have been observed in *G. max* and *G. soja* accessions. The second band only has been observed in isolated mitochondrial fractions. We have observed this second band in preparations from only a few samples so we do not know if variant forms exist in the population. All 235 *G. max* and *G. soja* accessions that we have tested showed the same GOT zymograms with respect to bands 1 and 3. Band 3 was found to be associated with green tissues or tissues which can become green, and with crude chloroplast isolations in preliminary fractionation studies. This would suggest that GOT band 3's subcellular activity site is in the chloroplast. Band 1 appeared to be found in the cytoplasm. Cytoplasmic- and chloroplast-associated GOT bands have been reported in other plants as well (Weeden and Gottlieb, 1980). The distinct subcellular origins as well as differences in tissue distribution strongly suggest that each observed GOT band was the product of unique loci.

C) Diaphorase (Dia): The diaphorases are a ubiquitous class of enzymes capable of reducing certain dyes (2, 6-dichlorophenol indophenol). In soybeans as well as other plants several molecularly different enzymes probably fall into the diaphorase activity group. In *G. max* and *G. soja* we have observed a total of 12 anodal Dia bands with accessions falling into one of 5 zymogram types (Fig. 1D). These 5 types were delineated by three



different electrophoretic variants. The first variant affects the expression of a 5-band cluster of bands close to the origin, with some accessions having all 5 bands and others lacking the fifth band while showing weaker third and fourth bands. The difference seems to be controlled by a single locus which has a dominant functional allele that expresses all 5 bands, and a recessive null (or weak) allele which shows the lesser pattern (Table 2A). The *Glycine clandestina* accessions examined to date were found to be polymorphic for a variant affecting the mobilities of just bands 1-4 in this cluster. These variants would suggest that in the *Glycine* species tested this 5-band Dia cluster is controlled by two loci, with the middle bands probably being heterotetramers and bands 1 and 5 homotetramers from these two loci. The second variant observed in *G. max* and *G. soja* accessions affects the mobilities of the seventh and eighth bands, with some accessions having fast migrating bands and others slow migrating bands. A single locus with two codominant alleles appears to control the difference (Table 2B). The third variant observed in *G. max* and *G. soja* accessions concerned the presence or absence of the tenth Dia band, but we have no inheritance data for this variant.

D) Malate dehydrogenase (MDH): 125+ *G. soja* and *G. max* accessions have been examined for MDH activity. All showed the same zymogram pattern. Subcellular fractionation studies revealed that at least 2 subcellular classes of MDH bands were included, mitochondrial and cytoplasmic.

Table 1. LAP band 1 inheritance data

Cross	Frequencies of genotypes in:					
	F <sub>2</sub>			F <sub>3</sub> segregating families		
	<u>FF</u>	<u>FS</u>	<u>SS</u>	<u>FF</u>	<u>FS</u>	<u>SS</u>
A73-25050 x PI 407.195 (type F x type S)	5	5	2	24	55	21
Amsoy x Wilson (type F x type S)	10	24	13			
Total: (observed)	15	29	15	24	55	21
$\chi^2$ (1:2:1) =	.02 = n.s.			1.16 = n.s.		

Table 2. Diaphorase inheritance data

Cross	Frequencies of genotypes in:					
	F <sub>2</sub>	F <sub>3</sub> segregating families				
<u>Part A. Dia variant 1</u>						
	<u>type 1</u>	<u>type 2</u>	<u>type 1</u>	<u>type 2</u>		
Cayuga x Mandarin (type 2 x type 1)	34	14				
Cayuga x Evans (type 2 x type 1)	17	7				
Lindarin x Norredo (type 1 x type 2)	45	20				
A73-25050 x PI 407.195 (type 1 x type 4)	3	0	81	33		
Altona x Chestnut (type 2 x type 1)	4	0	32	10		
Total: (observed)	103	41	113	43		
X <sup>2</sup> (3:1) =	.92 = n.s.		.55 = n.s.			
<u>Part B. Dia variant 2</u>						
	<u>FF</u>	<u>FS</u>	<u>SS</u>	<u>FF</u>	<u>FS</u>	<u>SS</u>
A73-25050 x PI 407.195 (type 1 x type 4)	1	1	1	14	32	16
Amsoy x Wilson (type 1 x type 3)	14	22	12			
Total: (observed)	15	23	13	14	32	16
X <sup>2</sup> (1:2:1) =	1.05 = n.s.			.2 = n.s.		

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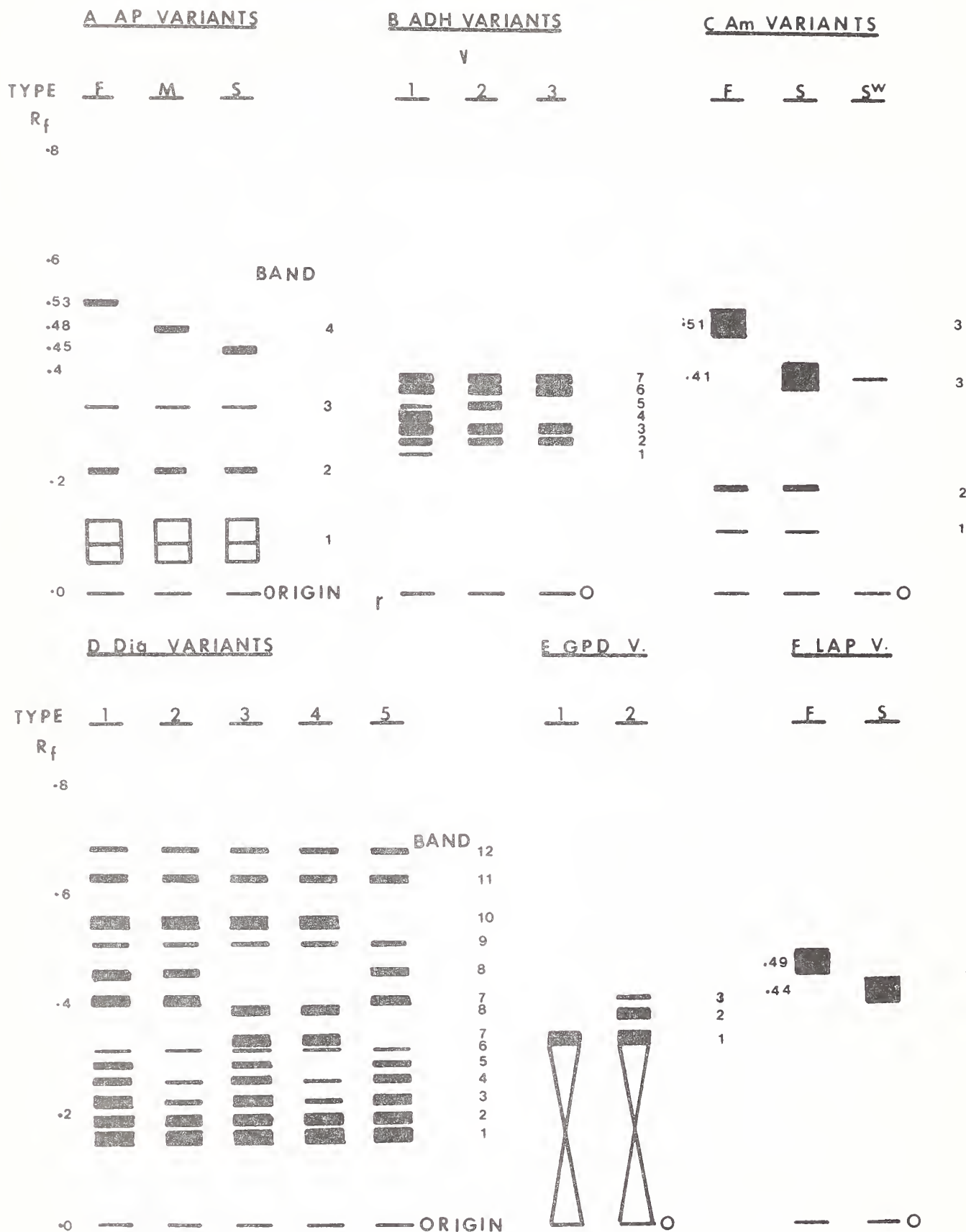
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2) <sup>245</sup> Electrophoretic classification of the early maturity groups of named soybean cultivars [-3]

Over the last several years, our lab has been collecting electrophoretic data for several [enzyme systems] in *G. max* and *G. soja*. While we have not yet completed analysis of all the available *G. max* or *G. soja* accessions, we have completed electrophoretic profiles for most of the named soybean cultivars in the early maturity groups (000-IV). We have been interested in using these electrophoretic profiles for cultivar identification, since a fairly complete identification can be made (Gorman and Kiang, 1977; and an article in preparation). Listed in the following table are the electrophoretic profiles on 12 enzyme systems for all of the varieties we have scored. Some of the enzyme types (Ep, Am, and AP) have been previously reported in this newsletter, and the Ep types were first observed and described by Buttery and Buzzell (1968). We included these enzymes in the table to make a more complete classification. Each classification in the body of the table represents a minimum of 5 electrophoretic observations. The zymogram types listed in the body of the table are those classifications described by Gorman and Kiang (1977), Gorman et al. (n.d.), and pictured in Figure 1. The columns in the table each represent an enzyme (ADH = alcohol dehydrogenase, Am = amylase, TO = tetrazolium oxidase, Ap = acid phosphatase, LAP = leucine amino peptidase, PGD = 6-phosphogluconate dehydrogenase, GPD = glucose-6-phosphate dehydrogenase, PGM = phosphoglucomutase, Ep = peroxidase, Dia = diaphorase, MPI = mannose-6-phosphate isomerase, and IDH = isocitrate dehydrogenase). Several of the enzymes (ADH, TO, Dia, and IDH) include more than one variable locus and all the zymograms, except MPI, represent the products of more than one locus. Genetic observations concerning these enzyme systems can be found in Gorman and Kiang (1978); Kiang (1981); Gorman et al. (n.d.); and Hildebrand et al. (1980), as well as part 1 of this newsletter. Listed at the end of the table are summaries of the number of varieties in each enzyme type, the total number of seeds scored per enzyme, the number of heterozygous seeds observed, and the total number of atypical seeds observed. We considered a variety to be of mixed (polymorphic) electrophoretic type when several seeds from that variety were scored into 2 or more enzyme types. This occurred in one of two ways; either the seed source (from R. L. Bernard at the U.S. Regional Soybean Lab at Urbana, IL) was mixed for two or more types, in which case the types are listed with commas, or seed batches obtained on different occasions had different enzyme types, in which case the types are listed with a slash between them. When only 1 or 2 seeds from a variety were found to be different from the large majority of seeds in that variety for a given enzyme type, they were counted as atypical seeds. Many of the atypical seeds were probably the result of experimental error; either by mixing seeds up, or by misreading zymograms. We hope that the table, along with the listed references and Figure 1, will be fairly self explanatory and will be of use to other soybean genetics laboratories or as an aid in cultivar identification.

# FIGURE 1. OBSERVED ELECTROPHORETIC ZYMOGRAMS



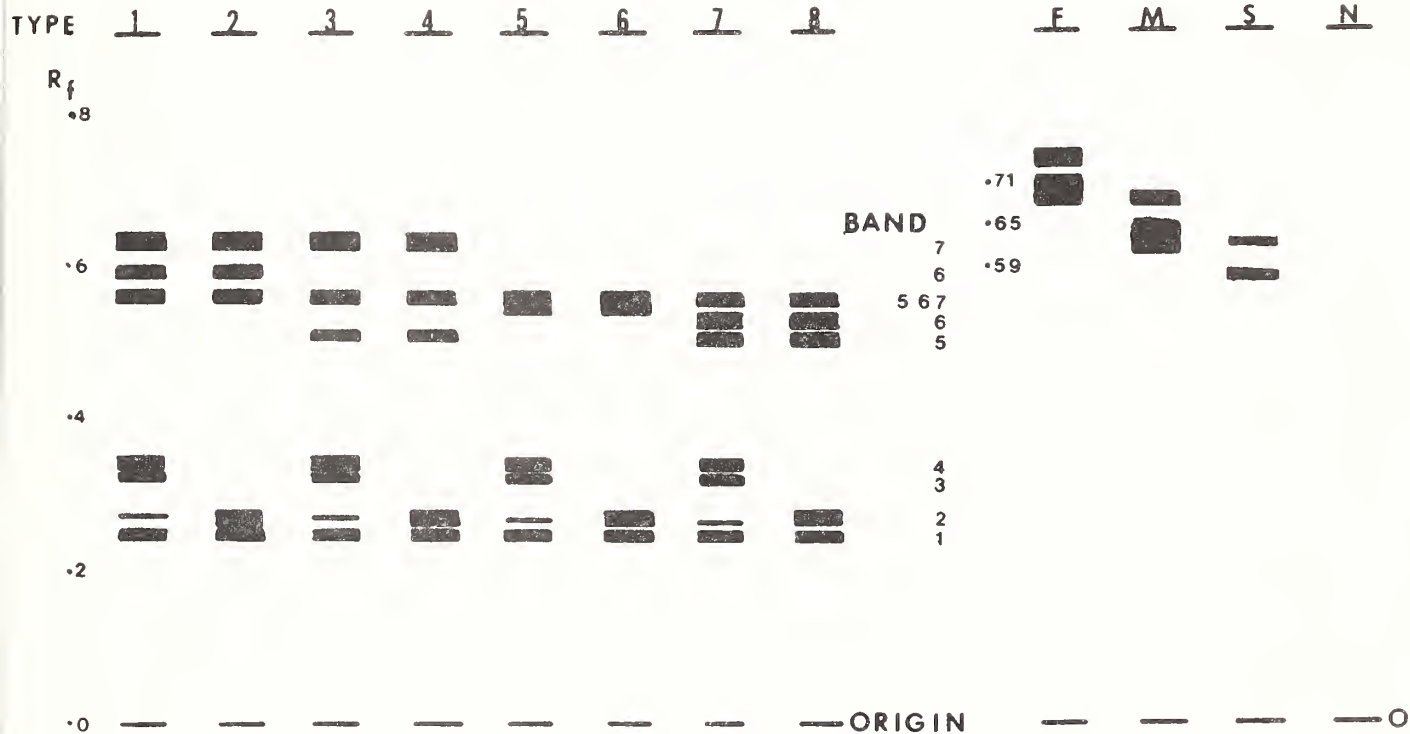
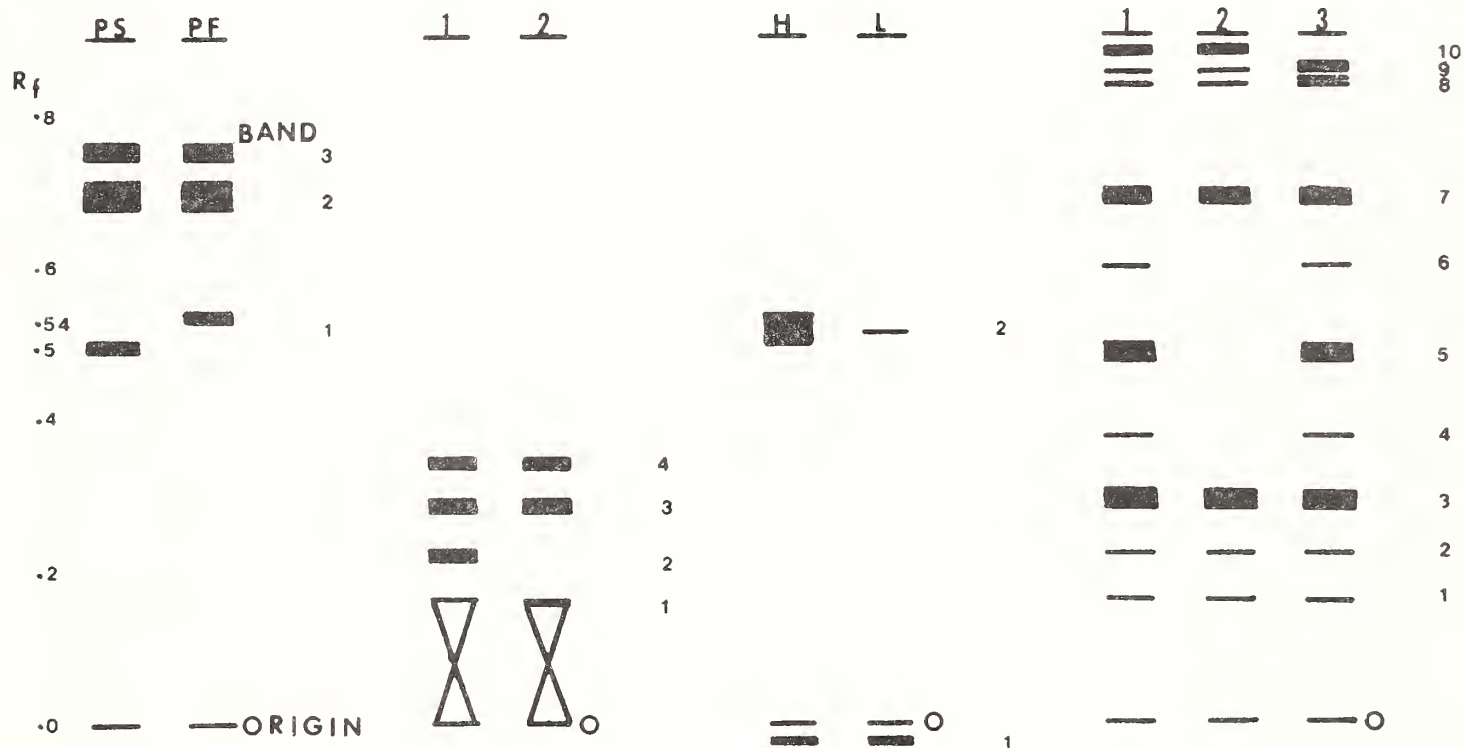
G IDH VARIANTSH MPI VARIANTSI PGM V.J PGD V.K EP VAR.L TO VARIANTS



Figure 1: Observed electrophoretic zymograms: A - fast, medium, and slow AP variants; B - types 1, 2 and 3 ADH variants; C - fast, slow, and weak slow Am variants; D - types 1-5 Dia variants; E - types 1 and 2 GPD variants; F - fast and slow LAP variants; G - Types 1-8 IDH variants; H - fast, medium, slow, and null MPI variants; I - types PS and PF PGM variants; J - types 1 and 2 PGD variants; K - high and low Ep variants; L - types 1-3 TO variants. Zymograms are drawn with the thickness of a band relative to its strength and the bands Rf position inversely relative to methyl blue. These zymograms are representative of zymograms observed from cotyledon samples within 24 hours from when seeds began to soak in water.

VARIETY ZYMOGRAM PROFILES

Zymogram Types

Variety / Enzymes:	ADH	Am	TO	AP	LAP	PGD	GPD	PGM	Ep	Dia	MPI	IDH
A-100	2	F	1	M	F	1	1	PS	L	1	M	2
Acme	1	S	1	M	F,S	1	2	PF	H	2	M	7
Ada	1	S	2	M	F	1	2	PS	L	1	M	1
Adams	2	F	1	M	F	1	1	PF	H	2	M	5
Adelphia	2	F	1	M	F	2	1	PF	H	2	M	5
Agate	1	F	3	M	F	2	1	PS	L	1	M	5
AK (FC 30.761)	1	F	1*	M	F	1	1	PS	H	2	M	5
Aksarben	1	F	1	M	F	1	2	PF	H	1	S	1
Altona	1	F,N	1	M	F	1	1	PS	H	2	F	5
Ancor	1	S	1*	M	F	1	2	PS	H	1	M	8
Amsoy + Amsoy 71	1	S	1	M	F	1	2	PF	H	1	M	8
Anoka	1	F	1*	M	F	1	1	PS	L	1	F	2
Aoda	1	F	1*	M	F	2	2	PF	L	1	F	7
Bansei	1	F	1	M	F	2	2	PF	L	1	F	7
Bavender Special A	1	F	1*	F	F	1	2	PS	L	2	M	8
Bavender Special B	1	F	1*	F	F	1	2	PS	H	2	M	6
Bavender Special C	1	F	1*	F	F	1	1	PS	L	2	M	8
Beeson	1	S	1	M	F	1	1	PS	L	2	M	2
Beeson 80	1	S	1*	M	F	1	1	PS	L	2	M	2
Bethel	2	S	1*	M	F	1	1	PS	L	1	S	8
Blackeye	1	F	1*	M	F	1	2	PF	H	1	F	7
Black Eyebrow	1	F	1	M	F	1	1	PS	H	2	M	5
Blackhawk	1	F	2	M	F	1	1	PS	L	1	M	1
Bombay	1	F	1*	M	F	1	1	PF	H	1	F	1
Bonus	1	S	1	M	F	1	1	PS	L	2	M	4
Boone	2	F	1*	M	F	1	1	PS	L	2	M	6
BSR 301	1	F	1*	M	F	1	1	PS	L	1	M	5

Variety / Enzymes:	ADH	Am	TO	AP	LAP	PGD	GPD	PGM	Ep	Dia	MPI	IDH
BSR 302	1	F	1*	M	F	1	1	PS	L	2	M	2
Burwell	1	F	1	M	F	1	1	PF	L	1	F	8
Calland	1	F	1	M	F	2	1	PS	L	2	M	2
Capital	1	F	1	M	F	1	1	PS	H	2	F	6
Carlin	1	F	1*	M	F	1	1	PS	H	2	M	6
Cayuga	3	F	1	M	F	1	1	PS	H	2	M	7
Century	1	F	1*	M	F	1	1	PS	L	2	M	2
Chestnut	1	S <sup>w</sup>	1	M	F	1	1	PS	H	1	F	2
Chief	1	F	1	M	F	1	1	PS	L	2	M	5
Chippewa	1	F	1	M	F	1	1	PS	L	2	M	2
Chusei	1	F	1*	M	F	2	1	PS	L	3	F	7
Clark	1	F	1	M	F	1	1	PS	L	1	M	6
Clay	1	F	1	M	F	1	1	PS	L	2	M	2
Cloud	1	F	1	M	F	1	1	PS	H	2	M	4
Coles	1	F	1*	M	F	1	1	PF	H	1	F	6
Columbia	1	F	1*	M	F	1	1	PS	L	1	S	1
Comet	1	S	1	M	F	2	2	PF	H	1,2	M,F	4
Corsoy	1	F	1	M	F	1	2	PS	H	2	F	6
Corsoy 79	1	F	1*	M	F	1	2	PS	H	2	F	6
Crawford	1	F	1*	M	F	1	1	PS	H,L	1	M	5
Crest	1	S	1	M	F	1	1	PF	H	1	M	3
Cumberland	1	F	1*	M	F	1	2	PS	H	1	M	5
Custer	1	F	1*	M	F	1	2	PF	L	1	M	5,7
Cutler	1	F	1	M	F	1	1	PS	L	2	M	5
Cutler 71	1	F	1*	M	F	1	1	PS	L,H	2	M	5
Cypress No. 1	1	F	1*	M	F	1	1	PF	H	1	F	4
Delmar	2	F	1	M	F	1	1	PS	L	1	S	6
Desoto	1	F	1*	M	F	1	1	PS	L	1	M	5
Disoy	1	F	1	M	F	1	1	PS	L	1	F	6

Variety / Enzymes:	ADH	Am	TO	AP	LAP	PGD	GPD	PGM	Ep	Dia	MPI	IDH
Douglas	1	F	1*	M	F	1	1	PS	L	2	M	6
Dunfield	1	F	1	M	F	1	1	PF	L	2	M	5
Dunn	1	F	2	M	F	1	1	PS	L	2	M	6
Earlyana	1	F	1	F	F	1	1	PS	L	2	N	8
Early White Eyebrow	1	S	1*	F	F	1	2	PF	H	2	F	4
Ebony	1	S	1	S	F	1	1	PS	L	2	M	7
Elf	1	F	1*	M	F	1	1	PS	L	1	M	7
Elton	1	F	1	M	F	1	1	PF	H	5	F	4
Emerald	1	F	1*	M	F	1	1	PS	H	2	M	3
Emperor	1	F	1*	M	F	2	2	PS	L	1	M	7
Ennis I	1	F	1*	M	F	1	1	PS	L	2	M	6
Etum	1	F	1*	M	F	2	2	PF	L	1	F	7
Evans	1	S	2	M	F	1	2	PS	H	1	F,M	1
Fabulin	2	F	1*	M	F	1	1	PF	L	2	F	6
Fayette	1	F	1*	M	F	1	1	PS	H	1,3	M	7
Flambeau	1	F	1	M	F	1	1	PF	H	2	M	6
Ford	1	F	1	M	F	1	1	PS	L	2	M	6
Franklin	1	F	1*	M	F	1	2	PS	L	1	M	7
Fuji	1	F	1	M	F	1	1	PS	L	1	F	7
Funk Delicious	1	F	1*	M	F	2	2	PS	L	1	M	7
Funman	1	F	1*	F	F	1	1	PF	L	2	M	8
Giant Green	1	F	3	M	F	2	2	PS	L	1	F	7
Gibson	1	F	1	M	F	1	1	PF	L	2	M	6
Gnome	1	F	1*	M	F	1	1,2	PS	L	1	M	5
Goku	1	F	1*	M	F	2	2	PF	L	2	M	7
Goldsoy	1	F	1	M	F	1	2	PF	L	1	F	4
Grande	1	F	1*	M	F	1	1	PS	L	1	F	5
Granger	1	F	1	M	F	1	1	PS	L	2	M	8
Grant	3	F	2	M	F	1	1	PS	L	2	M	6
Green and Black	3	F	1	M	F	1	1	PS	L	2	F	8

Variety / Enzymes:	ADH	Am	TO	AP	LAP	PGD	GPD	PGM	Ep	Dia	MPI	IDH
Guelph	1	F	1*	M	F	1	1	PS	H	2	M	2
Habaro	1	F	1	M	F	1	1	PF	H	1	F	4
Hahto (Mich.)	1	F	1	F	F	2	1	PS	L	1	F	6
Hakote	1	F	3	M	F	1	1	PS	L	1	F	7
Harbinsoy	1	F	1*	M	F	1	1	PS	H	2	S	6
Harcor	1	F	1*	M	F	1	2	PS	H	2	F	6
Hardin	1	S	1*	M	F	1	2	PS	L,H	2	F	6
Hardome	1	S	1	M	F	1	2	PF	H	2	M	4
Hark	1	F	1	M	F	1	1	PF	H	1	F	6
Harlon	1	F	2	M	F	1	1	PS	L	1	F	3
Harly	2	F	1	M	F	1	1	PS	H	2	M	1
Harman	1	F/S	1*	M	F	1	1	PF	H	2	F	6
Harosoy	1	S	1	M	F	1	2	PS	H	1	M	7
Harwood	1	S	1*	M	F	1	1,2	PF	H	1	M	8
Hawkeye	1	F	1	M	F	1	1	PF	L	1	M	1,5
Henry	1	S	1	M	F	1	1	PF	H	1	M	1
Hidatsa	1	F	1	M	F	2	1	PF	L	2	M	7
Higan	1	F	1	M	F	2	1	PS	L	3	F	5
Hobbit	1	F	1*	M	F	2	2	PS	L	1	M	5
Hodgson	1		1*	M	F	1	2	PF	H	1	F	6
Hodgson 78	1	F	1*	M	F	1	2	PF	H	2	F	6
Hokkaido	1	F	1*	M	F	2	1	PS	L	1	M	7
Hongkong	1	F	1*	M	F	1	2	PS	H	1	M	1
Hoosier	1	F	1	F	F	1	1	PS	H	2	F	6
Hurrelbrink	1	F	1*	M	F	2	1	PF	L	1	M	7
Illington	1	F	1	M	F	1	1	PS	L	2	F	7
Illini	1,2	F	1	M	F	1	1	PF	H	2	M	5
Ilsoy	1	F	1*	M	F	1	1	PS	L	2	M	4
Imperial	1	F	1	M	F	1	2	PS	L	1	M	7



Variety / Enzymes:	ADH	Am	TO	AP	LAP	PGD	GPD	PGM	Ep	Dia	MPI	IDH
Jefferson	3	F	1*	M	F	1	2	PS	L	1	M	7
Jogun	1	F	3	M	F	2	2	PF	L	1	F	8
Kabott	1	F	1	M	F	1	1	PF	L	1	F	8
Kagon	1	F	1	M	F	1	2	PF	H	1	F	4
Kahala	1	F	1*	M	F	1,2	1,2	PS	L	1	F	8
Kailua	2	F	1*	M	F	2	1	PS	L	1	F	8
Kanrich	1	F	1	M	F	2	1	PS	L	1	F	3
Kanro	1	F	1*	M	F	2	2	PF	L	1	M	7
Kanum	1	F	1	M	F	2	2	PF	L	1	F	5
Kent	1	F	1	M	F	2	1	PS	H	2	M	6
Kim	1	F	1*	M	F	2	1	PF	L	1	F	7
Kingston	1	F	1	M	F	2	1	PF	H	4	M	3
Kingwa	1	F	1	M	S	1	1	PS	H	2	S	1
Korean	2	F	1*	M	F	1	1	PS	H	1	F	3
Kura	1	F	1*	M	F	1	1	PF	L	1	F	8
Lakota	1	F	1*	M	F	1	1	PS	H	2	M	1
Lawrence	1	F	1*	M	F	1	1	PS	L	2	M	2
Lincoln	2	F	1	M	F	1	1	PS	L	2	M	6
Lindarin	2	S	1	M	F	1,2	2	PS	L	1	M	2
Lindarin 63	2	S	1*	M	F	1	2	PS	L	1	M	2
Linman 533	1	F	1*	F	F	1	1	PF	L	2	M	6
Little Wonder	1	F	1	M	F	1	2	PF	L	1	M	2
Macoupin	2	S	1	M	F	1	1	PF	L	1	M	5
Madison	2	F	1*	M	F	1	2	PF	L	2	S	6
Magna	1	S/F	1	M	F	1	2	PF	H/L	1	F	7
Manchu	1	F	1	F	F	1	1	PF	L	2	M	8
Manchu (Hudson)	1	F	1	F	F	1	1	PF	L	2	M	6
Manchu (Montreal)	1	F	1*	F	F	1	1	PS	H	1	M	8
Manchukota	1	F	1*	F	F	1	1	PS	H	2	M	6

Variety / Enzymes:	ADH	Am	TO	AP	LAP	PGD	GPD	PGM	Ep	Dia	MPI	IDH
Manchuria	1	F	1	M	F	1	1	PS	H	1	F	3
Mandarin	1	S	1	M	F	1	2	PF	H	1	F	4
Mandarin Ottawa	1	S	1	M	F	1	2	PF	H	1	F	4
Mandell	1	F	1	F	F	1	1	PS	L	2	M	8
Manitoba Brown	1	F	1	M	F	2	1	PS	L	2	M	5
Mansoy	1	F	1	F	F	1	1	PS	L	2	M	2
Maple Arrow	1	F	1*	F	F	1	2	PF	L	1	M	6
Maple Presto	1	S	1*	M	F	1	2	PS	H	2	F	3
Marion	1	S	1*	M	F	1	2	PF	H	1	F	5
McCall	1	F	1*	M	F	1	2	PF	H	1	M	3
Mead	1	S	1*	M	F	1	1	PS	L	2	M	3,7
Medium Green	1	F	1	M	F	1	1	PF	H	2	F	7
Mendota	1	F	1	M	F	2	2	PF	L	2	F	5
Merit	1	F/S	2	M	F	1	1	PS	L	1	M	1
Midwest	3	F	1	M	F	1	1	PS	L	2	M	2
Miles	1	F	1*	M	F	1	1	PF	L	1	M	5
Miller 67	1	F	1*	M	F	1	1	PS	H	1	M	4
Mingo	1	F	1*	F	F	1	1	PF	L	2	M	6
Minsoy	1	F	1	M	F	1	2	PS	L	4	M	3
Mokapu Summer	1	F	1*	M	F	2	2	PF	L	1	M	7
Monroe	1	S	1	M	F	1	1	PF	H	1	F	5
Morse	1	F	1	M	F	1	1	PF	H	2	S	3
Morsoy	1	F	1*	M	F	1	2	PF/PS	L	2	M	5
Mukden	1	F	2	M	F	1	1	PF	L	1	M	5
Nebsoy	1	F	1*	M	F	1	1	PS	H,L	2	M	7,5
Norchief	1	F	1	M	F	1	1	PF	H	2	M	6
Norman	1	S	1	M	F	1	2	PF	H	2	M	4
Norredo	1	F	1*	M	S	1	1	PS		2	M	1
Norsoy	1	F	1	M	F	1	1	PF	H	2	F	4
OAC 211	1	F	1*	M	F	1	2	PF	H	1	F	4

Variety / Enzymes:	ADH	Am	TO	AP	LAP	PGD	GPD	PGM	Ep	Dia	MPI	IDH
Oakland	1	F	1*	M	F	1	1	PS	L	2	M	1
Ogemaw	1	F	1	M	F	2	1	PS	L	2	M	5
Oksoy	1	F	1*	M	F	1	2	PF	L	1	M	5
Ontario	2	F	1*	F	F	1	1	PF	L	1	M	5
Osaya	1	F	1	M	F	2	1	PS	L	3	F	7
Ottawa	1	S	2	M	F	1	1	PS	L	1	M	1
Pagoda	1	F	1*	M	F	2	2	PF	L	2	M	8
Pando	1	F	1	M	F	2	1	PF	L	2	M	8
Patterson	1	F	1*	F	F	1	1	PF	H	2	M	6
Peking	1	F	1	M	F	1	1	PF	H	2	M	1
Pella	1	F	1*	M	F	2	1	PS	L	2	M	1
Perry	1	F	1	M	F	1	1	PS	L	1	S	5
Pennsoy	1	F	1*	F	F	1	2	PS	L	2	M	8
Pixie	1	F	1*	M	F	1	2	PS	L	1	M	7
Poland Yellow	1	F	1	M	F	1	2	PF	L	1	F	4
Polysoy	1	F	3	M	F	1	1	PS	L	2	M	2
Pomona	1	F	1*	M	F	2	1	PS	H	2	F	6
Portage	1	S	1	M	F	1	2	PF	H	1	M,F	3,7
Portugal	1	F	1*	M	F	2	1	PF	L	2	F	7
Pridesoy 57	1	F	1*	M	F	1	1	PF	H	2	M	4
Prize	1	F	1	M	F	1	2	PF	L	1	F	8
Protana	2	F	2	M	F	1	1	PF	L	2	M	6
Provar	1	F	1*	M	F	1	1	PF	H	1	M	6
Rampage	1	F	1*	M	F	1	1	PS	L	1	M	2
Renville	2	F	1*	M	F	1	1	PS	L	2	M	2
Richland	1	F	1	M	F	1	1	PS	L	1	M	1
Roe	2	F	1*	M	F	1	1	PF	L	1	M	5
Ross	2	F	1	M	F	1	1	PS	H	1	M	6
Sac	1	F	1*	M	F	2	2	PS	L	1	F	7
Sanga	1	F	1	M	F	1	2	PS	H	4	M	7

Variety / Enzymes:	ADH	Am	TO	AP	LAP	PGD	GPD	PGM	Ep	Dia	MPI	IDH
Sato-3	1	F	1*	M	F	1	1	PS	L	1	F	7
Scioto	1	F	1*	F	F	1	1	PS	L	2	M	2
Scott	1,2	F	1*	M	F	1	2	PF	L	1	M	5,1,3,7
Seneca	3	F	2	M	F	1	1	PS	L	2	M	5
Shelby	1	F	1*	M	F	1	1	PS	L	2	M	8
Shingto	1	F	1*	F	F	1	1	PF	H	2	M	5
Shiro	1	F	1*	M	F	1	1	PS	L	2	F	5
Sioux	1	F	1*	M	F	2	1	PF	L	2	M	8
Sloan	1	F	1*	M	F	1	2	PS	L	1	F	1
Sooty	1	F	1*	S	F	1	1	PS	L	2	M	1
Sousei	1	F	1*	M	F	2	1	PF	L	2	M	7
Soysota	1	F	3	M	F	1	1	PS	L	1	M	3
Sprite	1	F	1*	M	F	1	2	PS	H	1	M	5
Steele	1	S	2	M	F	1	1	PF	L	1	M	4
Swift	2	F	1*	M	F	1	1	PS	H	2	F	6
Tastee	1	F	1*	M	F	2	1	PF	L	1	F	5
Toku	1	F	3	S	F	2	1	PF	L	1	F	7
Tortoise Egg	1	F	3	M	F	1	1	PS	L	1	M	7
Traverse	1	F	1*	M	F	1	2	PF	H	2	M	6,2
Troy	1	F	1*	F	F	2	2	PS	H	1	M	2
Union	1	F	1*	M	F	1	1	PS	H	1	M	5
Vansoy	2	S	1*	F	F	1,2	1,2	PF	H	2	F	2
Verde	1	F	1*	M	S	2	2	PF	L	1	F	8
Vickery	1	F	1*	M	F	1	2	PS	H	2	F	6
Viking	1	F	1*	M	F	1	1	PF	H	2	M	8
Vinton	1	F	1*	M	F	1	1	PF	H	1	M	6
Vinton 81	1	F	1*	M	F	1	1	PF	H	1	M	8
Virginia	1	F	1	M	F	1	1	PS	L	2	M	3
Wabash	1	F	1*	M	F	1	1	PS	L	2	M	5
Waseda	1	F	1*	M	F	1	2	PF	L	1	F	7

Variety / Enzymes	ADH	Am	TO	AP	LAP	PGD	GPD	PGM	Ep	Dia	MPI	IDH
Wayne	1	F	1	M	F	1	1	PS	L	2	M	7
Wea	1	F	1*	M	F	1	1	PF	L	2	M	5
Weber	1	F	1*	M	F	1	1	PS	H	2	F	6
Wells	1	S	1*	M	F	1	1	PS	L	2	M	8
Wells-2	1	S	1*	M	F	1	1	PS	L	2	M	8
Wilkin	1	F	2	M		1	1	PS	L	1	F	7
Will	1	F	1*	M	F	1	1	PS	H,L	1	M	5
Williams	1	F	1*	M	F	1	1	PS	H	1	M	5
Williams 79	1	F	1*	M	F	1	1	PS	H	1	M	5
Williams 82	1	F	1*	M	F	1	1	PS	H	1	M	5
Willomi	1	F	1*	M	F	2	2	PF	L	1	F	7
Wilson	1	F	1*	M	S	1	1	PS	L	3	S	1
Wing Jet	1	F	1*	M	F	1	1	PS	L	2	M	7
Wirth	1	F	1*	M	F	1	1	PS	L	2	M	2
Wisconsin Black	1	F	1*	F	F	1	1	PS	H	2	M	4
Wolverine	1	F	1*	M	F	2	1	PF	L	1	M	5
Woodworth	1	F	1*	M	F	1	1	PS	L	2	M	5
Wye	3	F	1*	M	F	1	1	PF	L	2	M	5
Yellow Marvel	1	F	3	M	F	1	1	PF	L	2	M	7

\*These TO's were not scored for the second TO variant and therefore some might be type 3.

[illegible]

Variety / Enzymes:	ADH	Am	TO	AP	LAP	PGD	GPD	PGM	Ep	Dia	MPI	IDH
Zymogram type totals:												44
(continued)												(7)
												30
												(8)
Total mixed varieties:	2	4	0	0	1	3	4	1	6	2	3	7
Total No. of seeds scored:	2605		1753		2398		1650		2429		1638	
		3107		1534		1712		1658		1704		1833
Total No. of atypical seeds:	4	5	2	0	0	8	4	2	5	0	4	18
No. of heterozygous seeds:	-	9	1	2	1	-	-	0	-	0	1	5

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15245 A somatic approach to soybean genetics

For the past five years, our laboratory has been attempting to establish a system of somatic cell genetics for soybean. Although our results are far from complete, they are sufficiently encouraging to suggest that such a system will be practical and that it should be possible to construct a complete genetic map within a few years. The rapidity with which this can be done relies in part on the somatic genetics discussed in this report and in part on a molecular genetic analysis which is now underway. This communication is a summary. Details have been published already, are available upon request, or will be published in full length in the near future.

Using techniques established by Gamborg and others (Gamborg and Wetter, 1975), it is possible to grow most varieties of soybean in cell culture. In general, it takes 2-4 weeks to establish healthy callus culture from seedling roots or hypocotyls and another 3-6 weeks to prepare suspension cultures. The latter are suspensions of small cell clumps which are maintained in an exponential phase of growth ( $5 \times 10^5$ - $2 \times 10^6$  cells/ml) by 1/4 dilution every 2-3 days. When grown at 33°C these cultures divide, on the average every 24 hrs (at 22°C division occurs every 48 hrs) (Chu and Lark, 1976). Below a titer of  $5 \times 10^4$  cells/ml soybean cultures fail to grow and eventually die. Above this titer, cells will survive and divide in defined minimal media. When cells become damaged or die, they eventually liberate toxic material which kills healthy cells present in the same culture. The cooperative requirement for some essential nutrient and the liberation of toxic material upon death make it difficult to obtain rare mutant cells by selective killing of parental wild type. To circumvent this, a plating technique was developed (Weber and Lark, 1979) which allows mutant cells to be maintained under selection while removing the toxic material liberated by dying parental cells. In this process a feeder culture system is maintained in which feeder cells are replaced every 2 days.

Using this system, it was possible to establish a quantitative system in which to study mutagenesis (Weber and Lark, 1980). The results of this study indicated that rates of inherited variation could be achieved in which variants (mutants) would occur in frequencies as high as 3 per  $10^4$  surviving cells. Excellent mutagenesis was obtained with MMS, NNG or U.V. irradiation. The worst mutagen was EMS.

Sibling selection of mutants: The high frequencies of variation obtained with U.V. encouraged us to try sib-selection. Haploid cells (see below) were irradiated to 1% survival and surviving clumps plated to obtain isolated callus clones. Each surviving callus was divided and one part tested selectively while the other was grown under nonselective conditions. Several thousand of these colonies were tested for auxotrophy, cold sensitivity, temperature sensitivity, and salt dependence-tolerance. Mutants were isolated in all classes. Details of the isolation have been published (Zhou et al., 1982) or will be published (Roth and Lark, Plant Cell Reports, in press). After the final isolation, the frequency of variants declined. We believe that this was due to contamination with wild-type parental cells which took over the growing culture. Stable variants which could be maintained nonselectively were isolated

with a frequency of ca. 1/5000. All of these have been grown continuously in cell culture for >200 generations. Improving the technique (Roth and Lark, Plant Cell Reports, in press) could, in the future, increase the frequency of variants.

Preservation of cell lines and clones: To isolate pure cell lines, it is necessary to prepare protoplasts and from these regenerate clones derived from single cells. To do this, one must prepare growing cell suspensions from callus. Because of the scale of selection, this is only feasible if one has ascertained which callus is mutant. Until recently, it was not possible to preserve cells in a nondividing state while testing for mutant phenotype. Recently, it has become possible to freeze soybean in liquid nitrogen and maintain viability (Weber, unpublished data; Roth and Lark, unpublished data). This allows a more efficient mutant screen and insures preservation of mutant genotypes. Details of this technique will be published after we have established the length of time for which cells can be stored.

Haploid and partial haploid lines: The studies on sibling selection outlined above were carried out using a haploid line of soybean (Weber and Lark, 1979) originally obtained as callus from Dr. Beversdorf. The callus had been prepared from a haploid plant which arose as the result of a cross between male-sterile lines. Eventually this line became polyploid. The ability to maintain cells in the frozen state should avoid a recurrence of such a loss. We are currently reconstructing haploid lines from plants obtained from male-sterile crosses. We also have obtained partial haploid lines by chemical treatments leading to misdivisions. This treatment is outlined below.

Induction of misdivisions by the herbicide CIPC: Previous experiments with algae had shown that the herbicides IPC and CIPC can induce abnormal division. We have used this treatment to induce loss of chromosomes in soybean. For these experiments we used a soybean suspension culture derived from heterozygous plants produced by crossing PI 290136 with 'Minsoy'. The  $F_1$  seeds were provided by Dr. R. Palmer. Each of the parent lines is homozygous recessive for different, unlinked, fluorescence markers [Minsoy: ( $fr_1fr_1Fr_2Fr_2$ ) PI 290136 ( $Fr_1Fr_1fr_2fr_2$ )]. Hence, each parent is nonfluorescent whereas the  $F_1$  heterozygote is fluorescent. Callus of the  $F_1$  heterozygote is fluorescent whereas calli from either parent are not. After treatment of the heterozygous suspension culture with CIPC (8 hrs  $7^\circ\text{C}$ ,  $1 \times 10^{-3}$  M CIPC 48 hrs  $7^\circ\text{C}$ ), we obtained about 10% surviving clones. Among these survivors 20% were nonfluorescent. Four of the surviving clones were passaged into suspension culture: three of these were fluorescent and one was nonfluorescent. All showed reduced numbers of chromosomes: 23-36.

To test whether the products of misdivision were capable of expressing auxotrophic mutants, we treated the four segregant suspension cultures with U.V. After recovery (ca. 1 week) we starved the cultures for ribonucleosides and then treated the cultures with 5-fluorouracil, an analog which is incorporated into RNA to produce inaccuracies capable of poisoning the cell. After 8 hr of growth in 5-fluorouracil, the cultures were plated on normal medium supplemented with ribonucleosides. Fifty clones were picked from each culture and tested for growth with or without nucleosides. Thirty one auxotrophs were detected, 14 in one segregant culture, 17 in another and none in the third and fourth. All of these results support the loss of chromosomes from cultures

treated with CIPC. In addition, the success of 5-fluorouracil selection demonstrates the possibility to select auxotrophs by use of inhibitors.

Potential for genetic mapping: The availability of mutants in partial haploid cell lines makes it possible to begin mapping genetic loci by fusion segregation tests. Recessive mutants which are expressed in partial haploids must belong to linkage groups limited to the chromosomes which are haploid. By fusing cells, one can produce somatic heterozygotes complementing the recessive mutation. Loss of the corresponding wild type chromosome will again lead to expression of the recessive phenotype. If the chromosomes which are lost can be differentiated, it becomes possible to assign each mutation to a chromosome. Presently, we are developing a molecular method for differentiating soybean chromosomes.

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1) <sup>45</sup> Protein content in grain and lysine content in protein of soybean mutants,  
induced by chemical mutagens and gamma rays

During the study of genetic activity of a number of chemical and physical mutagens about 1000 morphological mutants of different types were derived.

Such mutagens as nitrosomethyl urea (NMU), nitrosoethyl urea (NEU), ethylenimine (EI), ethylenoxide (EO), nitrosodimethyl urea (NDMU), dimethyl-sulfate (DMS), and gamma rays were used.

As the initial material for mutations induction, varieties 'Lanka', 'Hybrid 89-10', 'Kirovogradskaya 4', 'VNEEMK 9186' and 'Peremoga' were used. The above mentioned chemical mutagens were applied in water solutions, besides NMU, EO and DMS were used in gas phase also.

When the study of inheritance of changed characters was finished, the majority of mutants was rejected because they did not represent a significant interest for breeding.

The remainder are actually under trials for productivity in different breeding nurseries. These mutants are characterized as lodging resistant, early matured, have increased productivity and other valuable agricultural characteristics.

In this paper the results of grain protein content determination and the results of protein's lysine determination of  $M_5$  mutants are presented. Tables 1 and 2 demonstrate that several mutants outdid the regionally distributed variety 'Bucuria' in their data of evaluation. The initial varieties have less protein and lysine than Bucuria variety has. Significantly interesting are No. 271 and No. 521 mutants because of their protein content. During the period of 2 years, they outyielded the standard 4.20-4.55%. It is necessary to say that N 84 and N 251 mutants have the increased protein and lysine content simultaneously.

Thus, the results showed that among morphological mutants may occur the forms that are characterized by valuable chemical indices.

Table 1. Protein content of some soybean mutants

Mutant No.	Initial variety	Mutagen	Protein content (%)	
			1979	1980
-	Bucuria, standard	-	28.7	26.2
84	Kirovogradskaya 4	gamma-rays	33.4	27.8
92	Kirovogradskaya 4	NMU 0.00625%	31.7	27.8
137	Kirovogradskaya 4	gamma-rays	31.2	26.5
251	Hybrid 89-10	EI 0.01%	30.0	28.0



Table 1. *Continued*

Mutant No.	Initial variety	Mutagen	Protein content (%)	
			1979	1980
236	Lanka	NMU Ig 10 days	31.3	27.8
237	Lanka	NMU Ig 10 days	33.7	27.3
265	Peremoga	EO 0.05%	31.2	28.2
271	VNEEMK 9186	NDMU 0.025%	32.3	31.0
275	Kirovogradskaya 4	EO 10 drops	32.5	28.2
521	Peremoga	NEU 0.0125%	31.7	32.3

Table 2. Soybean mutants with increased lysine content in protein

Mutant No.	Initial variety	Mutagen	Protein content (%)	
			1979	1980
-	Bucuria, standard	-	8.23	7.84
3	VNEEMK 9186	gamma-rays	8.71	8.56
4	VNEEMK 9186	NMU Ig 5 days	8.38	8.54
8	Peremoga	gamma-rays	8.39	8.74
26	Peremoga	DMS 0.04%	8.78	8.46
84	Kirovogradskaya 4	gamma-rays	8.49	8.52
141	VNEEMK 9186	DMS 1ml 4 days	8.39	8.42
251	Hybrid 89-10	EI 0.01%	9.06	8.48
252	Peremoga	EI 0.02%	8.29	8.58
258	VNEEMK 9186	DMS 0.01%	8.25	8.46
272	Peremoga	DMS 1ml 4 days	8.38	8.76
276	VNEEMK 9186	NEU 0.0125%	9.22	8.46
328	Hybrid 89-10	gamma-rays	8.87	8.80
329	Kirovogradskaya 4	gamma-rays	8.87	8.40
706	Kirovogradskaya 4	NEU 0.05%	8.32	8.68

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2) <sup>1945</sup> The lectin content in different varieties of soybean in connection with improving nutritive qualities [ ].

The nutritional value of soybean protein is reduced in certain rate by inhibitors of tripsin, chemotripsin and lectins, too. Proteins that are capable of linking surface structures of erythrocytes and other type of cells to cause their agglutination, are called phytohemagglutinins or lectins. Moreover, they form complexes with many polysaccharides. The main part of lectins is located in grain of the major plant species, but they are present in ovary, style, stamens, roots and stems in negligible quantity (Golynskaya et al., 1975; Gatehouse and Boulter, 1980).

Lectins are characterized by a broad range of action on living cells, and on cells of human and animal gastrointestinal tract that cause reduction in nutritional matter absorption and may contribute to penetration of bacteria and toxins to blood (Liener, 1976; Pestrai et al., 1979). The matter of lectin's toxicity is most important because legumes and soybean especially occupy the main part of the daily livestock diet.

When soybean lectins were incorporated into rats' diet at a level equivalent to the content of them in raw soybean meal, a significant growth depression of rats was obtained (Liener, 1974). Liener concluded from his experiments that approximately one-half of the growth depression was obtained after feeding rats uncooked soybeans, due to these lectins. Another half of the growth inhibition was attributed to tripsin inhibitors. The heat treatment of soybean meal was used to destroy and inactivate the toxicity, but this process involved certain economic outlays and had other disadvantages (Dimov, 1979).

The fact that the majority of legumes possess 2-10% lectins of whole protein gives reason to consider that they play an important role in reducing nutritive qualities. What is more, there are some data indicating that lectins protect germinating legume seeds against soil microorganisms (Barkai-golan et al., 1978) and insects (Jansen et al., 1976).

It is known that soybean lectins consist of 4 glycoproteins, and the most of them are 120,000 (Catsimpoolas and Meyer, 1969; Lotan et al., 1974). Investigation of lectin content in 102 soybean varieties showed that 5 of them lacked this substance, and it was dependent on recessive gene *le le* (Pull et al., 1978; Orf et al., 1978).

The lectin content of different soybean seed samples was determined regarding the variety genotype and origin. These data are necessary for breeding to increase soybean nutritive quality.

As initial material 26 varieties of soybean were used. They originated from the USSR, USA and Europe. Seed meal was defatted in boiling chloroform, and then extracted in acid distilled water pH 4.5 at 37°C for 2 hr and at 4°C for 18 hr.

The mixture was centrifuged to remove plant debris. Agglutinated proteins were precipitated from extract by ethanol fractionation. The precipitates produced at 30% alcohol saturation were rejected. Protein precipitates produced at 30-50% alcohol saturation (the I fraction), at 50-76% (the II fraction) and at 30 to 76% (summarized), were collected by centrifugation at 6,000 G for 20 min and then lyophilized (Boyd and Shapleigh, 1954). The precipitates were dissolved in buffered salt solution at pH 7.2-7.4 with

appointed percentage for agglutination. Erythrocytes of the human blood group I were used for agglutinative activity determination. These erythrocytes were washed off in buffered solution and some of them were treated with trypsin (Lutsik et al., 1980). The agglutination reaction was made on glass slides (Dosse, 1969) and in test-tubes (Hable and Zel'tsman, 1972).

Determination of hemagglutinative depression percentage in carbohydrate presence (by Landshtayner) was made at its constant concentration and progressive reduction of lectin content. Depression percentage was calculated by formula:

$$x = \frac{\text{control number} - \text{trial number}}{\text{control number}} \times 100\%,$$

where trial number is number of the last test-tube where agglutination took place yet in carbohydrate presence. Number of control is number of the test-tube in which agglutination took place in carbohydrate absence.

The agglutinative protein activity of 26 soybean varieties was analyzed at 50% and 76% of alcohol saturation. It was determined that in different varieties the agglutinable proteins were present in different fractions. In most varieties they were present in the I fraction, and only a few varieties had protein in the II fraction. Basing on lectin localization in different fractions, all tested varieties can be divided into three groups. The first group contains 24 varieties, which have lectins in the I fraction; variety 'Belosnezshka' belongs to the second group and contains lectins in both fractions, and variety 'Lanka' belongs to the third group and contains lectins in the second fraction. It must be mentioned that the first group of varieties demonstrated agglutinative activity in the II protein fraction with trypsinized erythrocytes. That may be explained as traces of the basic fraction I, or as the presence of one of lectin isoforms, which has low agglutinative activity. Percentage of summarized fraction having agglutinative activity in unfatted meal changed from 1.42 to 4.17% depending on the variety (Table 1). The highest content of this substance was in varieties 'Terezinskaja 2', 'Khersonskaja 8', 'Severnaja 5', 'Panonia'. There are few lectins in seeds of varieties Belosnezshka, 'Krasnodarskaja 10', 'Saljut 216', 'Anoka'. The samples have the higher agglutinative titer if they have high content of lectins. The lectin titer was 1:4096-1:2.

Percentage of hemagglutinative depression by 0.005 M arabinose was studied for every variety (Table 1). American varieties were distinguished by these data, and they had depression percentage of about 50 and more.

Analyzed samples clearly divided into two groups of lectin quality, by eye. One group of varieties had "meal" type lectins which, after alcohol precipitation and two cleanings by ammonium sulfate, looked like a loose white flouring mass. The next varieties group had glass-like lectins as limpid sugar sand.

Division of pure lectins by disk-electrophoresis method made it possible to select a group of varieties which had similar electrophoregrams.

Analysis of agglutinative protein quantity and agglutinative titer regarding variety origin demonstrated high lectin content in European varieties, Ukrainian breeding especially.

American varieties had low lectin content, as did the Chinese variety 'Man-Tsan-Tsin'. Thus, results of the present investigation show significant differences in lectin content of soybean varieties that suggests successful breeding on this character.

Table 1. The total agglutinative proteins content of different soybean varieties

Variety	Origin	Agglutinin content, %	Titer	Depression of agglutination, %
Terezinskaja 2	Ukrainian SSR	4.17	1:4096	30
Kirovogradskaja 5	"	2.68	1:512	30
Khersonskaja 8	"	3.33	1:256	25
Lanka	"	2.72	1:128	0
Kirovogradskaja 3	"	2.33	1:128	14
Belosnezshka	"	1.42	1:64	17
Lumina	Moldavian SSR	2.67	1:512	30
VNEFMK 9186	RSFSR	2.17	1:512	20
Kujbishevskaja 77	"	1.87	1:64	33
Krasnodarskaja 10	"	1.68	1:32	20
Severnaja 5	"	3.73	1:256	12.5
Amurskaja 450	"	2.32	1:256	12.5
Saljut 216	"	1.75	1:16	20
Vengerskaja 48	Hungary	2.48	2:256	12.5
Caloria	GDR	2.37	1:128	25
Pavlikeni 2	Poland	2.12	1:128	25
Panonia	Rumania	3.77	1:1024	10
Man-Tsan-Tsin	China	1.87	1:2	100
Anoka	USA	1.46	1:128	27
Capital	"	2.33	1:64	33
Wirth	"	2.03	1:64	50
Hawkeye	"	2.02	1:32	67
Morsoy	"	2.27	1:16	60
Steele	"	2.08	1:16	50
Hodgson	"	1.87	1:16	100

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1) Decapitation technique to increase grain yield in soybean,

In soybean, pod clusters were generally formed at the nodes of stem and/or at those of branch, but at the higher nodes, pod clusters failed to develop sufficiently, pods and seeds seemed to be smaller and empty (unfilled). Then, if we can control its branching capability, and cause branching earlier and more numerous to bring out more flower and pod clusters at the nodes of mid-stem and those of mid-branch, it will be sure that flowering and/or pod formation occur at short interval of time and be numerous. In spring, soybean grown in well-amended soil, if subjected to decapitation technique when having 4-5 trifoliolate leaves, will begin branching earlier, branches may develop well and grain yield increases steadily. It is recommendable to use decapitation technique in soybean having 7-8 trifoliolate leaves in summer. When subjected to decapitation technique, soybeans flower 4-5 days earlier and interval of time of flowering is shorter. This gives more horizontal branches exposed to more sunlight; thus, soybeans may produce more filled pods and harvest can be done earlier because soybean matures more uniformly. Let us use decapitation technique in soybean at proper time; if earlier, plants are still weak and if later, branching becomes more numerous and then there is no action at all. When this decapitation technique is applied on a commercial scale, grain yields may differ from 14% to 22% when soybean is grown at proper time.

Table 1. Interaction of decapitation technique versus number of flowers, number of pods, pod weight and seed weight in spring soybean

Treatment	No. of flowers	No. of pods (%)	Pod weight (%)	Seed weight (%)
Without decapitation	88	100	100	100
With decapitation	123	130	130	130

\*Removed a terminal internode and uppermost leaf when having 5 trifoliolate leaves.

\*\*Flowering occurred at 76-80 days after sowing.

Table 2. Interaction of decapitation technique versus total length of branches per plant, leaf number per plant, number of flowers per plant and grain yield

Treatment stage	Total length of branches/ plant (cm)	No. of leaves per plant	No. of flowers per plant	Yield index (%)
5-leaf	129	65	359	82
6-leaf	---	---	---	---
7-leaf	191	109	460	154
8-leaf	---	---	---	---
9-leaf	179	87	438	142
Without decapitation	168	81	407	100

- NB. 1) Total length of branches is measured at 70 days after sowing  
 2) Number of leaves and number of flowers are counted at 80 days after sowing.  
 --- Data not computed.

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2) Nontillage is a technique in growing soybean (*Glycine max* (L.) Merrill)

Of course, the best type of soil for growing soybean (*Glycine max* (L.) Merrill) is a light textured soil, rich in humus, and its acidity is around neutral (not acid). Such a soil may be neither influenced by saline water nor under flooded condition. But in South Vietnam, especially in the Mekong Delta and in favorable weather conditions, soybean is able to grow and develop well and give a high seed yield on relatively heavy clay with pH over 4.5. There, soybean can be grown without land tillage; this has been clarified by experimental data in commercial scale.

This technique of nontillage is successfully applied both in sandy soil of island and in rice farmer field rich in humus.

The fact that soybean can be grown without land tillage on a variety of light textured soil as mentioned above is apparently accepted but there is also doubts for the feasibility of growing soybean in rice farmer field, of which, soil is quite heavy and more or less affected by ions  $Al^{+++}$  and/or  $Fe^{++}$  in dry season. But experimental data showed that there is no significant difference between two cultural practices with land tillage and nontillage. In fact, in many areas, when using proper cultural practices, nontillage soybean growing resulted in a seed yield of approximately 2-3 tons per ha.

In rice farmer field, growing soybean without land tillage means to seed or broadcast soybean seed right after rice harvest, when soil is still wet.



In floating rice area, soybean is directly seeded after rice harvest; then farmer does slightly tap on or use a long handled harrow to make soybean seed fall in wet soil surface. The remaining straw of floating rice is then served as a means of retaining soil moisture to meet water requirement of soybean for a long period of time. It was observed that when a crop of floating rice was successful, a crop of soybean followed was apparently successful, too.

In different rice fields, when soil was still wet, weeding was done after rice harvest and in better way, if there was much straw left, then the farmer made a so-called thin mat of straw stretched over the ground and burned it to kill pathogenic factors, weeds and also remaining straw. Besides, after burning, ash has an effect of neutralizing surface soil acidity and to provide some essential micro-elements to the plant. Afterwards, farmer digs small holes, put 2-3 soybean seeds in each hole, covers soybean seed with ash, and later stretches a mulch over the soybean field to maintain soil moisture.

In summary, in order to have good results from nontillage technique, we must pay attention to five main purposes following:

- Let's plant soybean when soil is still wet, not much dried out;
- After harvest, rice field must be free of weeds;
- In heavy soil, it is advisable to use a sharpened stick to dig a hole not large so that roots can easily develop well;
- After seeding, soybean seeds must be covered with ash or with ash mixed with different kinds of chemical fertilizers;
- In dry season, soil surface must be covered with a mulch, thick enough to retain soil moisture and also to prevent weeds.

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245  
Exotic soybean (*Glycine max* (L.) Merrill) observational yield performance trial in Kien Giang province - Mekong Delta \* Vietnam \* dry season 1981-1982 [ ]

In the light of mutual technical assistance, six soybean varieties, namely 'Bon minori' (2 lines), 'Enrei' (2 lines), 'Akiyoshi' and 'Hyuuga', were forwarded by registered mail from Japan to Vietnam in May, 1981. Due to its long postal course, soybean seed was only received in November 1981. A month later, seeds were planted on December 10, 1981, at the provincial seed farm of Kien Giang province and then harvested on February 26, 1982.

Materials and methods. The intent of experiment is firstly to determine the adaptability to local environmental conditions of six soybean varieties from Japan, newly introduced in Kien Giang province in November, 1981; secondly, to introduce to local farmers some new high-yielding soybean varieties, which are also resistant to common disease and insect pests; and, thirdly, to recommend growing soybean, a cash crop, in rice farmer's field in order to uncover the prospect of intensive cultivation and/or multiple cropping and also to amend soil fertility which is not fertile, pH  $5.5 \pm 0.5$  in the

quadrangle Long xuyen. Six soybean varieties with their respective entry number and number of seed planted were the following:

<u>Name</u>	<u>Entry number</u>	<u>Number of seed planted</u>
Enrei	040691	17
Akiyoshi	090125	34
Hyuuga	090204	29
Enrei	040697	15
Bon minori	040705	24
Bon minori	040207	17

Seeds were sown on the terrace recently built up in 1981 of Kien Giang provincial seed farm Service of Agriculture (20° 07' N), more or less representative of acid sulphate soil in the area. Seeding procedure was planting one seed per hill 2 cm deep, and spacings were 40 cm between rows, 10 cm between hills and 80 cm between varieties. The design was an observational yield performance trial without replicate; each variety was grown in a single row or in two subsequent rows, depending on number of seed in hand. Fertilizers were used with formula 40-80-30 kg/ha N, P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O and nursery soil was disinfected with Basudin 10G at the rate of 30 kg/ha.

Results and discussion. Based on experimental data, the first fifteen days after seeding, all six soybean varieties tested had a satisfactory germination and a good initial plant vigor, thanks to some late rains and a favorable soil moisture. But days after days, surface soil dried out quickly, and irrigation was then practiced with water stored in surrounding ditches, the only source of irrigation, more or less acid, which was apparently a limiting factor of plant vegetative growth. By the time, only Hyuuga continued to grow, while the other five soy varieties stopped growing and soon became stunted. In Hyuuga, plant height was 30 cm  $\pm$  0.5 and in others, plant height was approximately 15-20 cm.

In comparison of percent of field emergence and percent of surviving plants until harvest, in three varieties, Enrei (040691), Akiyoshi and Bon minori (040207), percent of surviving plant at harvest was very low, ranging from 10% to 21%, though their respective percent of field emergence seemed to be satisfactory. It was noticed that, partly, they were damaged by the common insects *Melanagromyza soja*, *Pseudaletia unipuncta* and *Cacoecia* sp.

In Hyuuga, Bon minori (040705) and Enrei (040697), their respective percent of surviving plant ranges from 47% to 64.5% (Table 1), the highest, but only Hyuuga gives a fair number of sound seed while the two other soybean varieties gave a lower number of seed and mostly seeds were small and wrinkled. Symptom of damage caused by *Maruca testutalis* was also noticed in Hyuuga soy variety.

At last, only Hyuuga soybean variety appeared to be the most promising one at this locality in dry season 1981-1981, but the observation above stated need to be understood with reserve. Consequently, this type of experiment should be repeated in coming rainy season 1982 for further study on the performance of these soybean varieties before reaching a definite significant reliable conclusion.

Table 1. Number of seed planted and percent of field emergence of six exotic soybean varieties tested in Kien Giang province - Mekong Delta - SR Vietnam, dry season 1981-1982.<sup>a</sup>

Variety name	Entry number	No. of seed planted	No. of seed germinated	No. of plants harvested	% of field emergence	% of surviving plants
Enrei	040691	19	15	2	78.9	10.5
Akiyoshi	090125	35	22	7	62.8	20.0
Hyuuga	090204	31	25	20	80.6	64.5
Enrei	040697	17	14	8	82.3	47.0
Bon minori	040705	26	20	14	76.9	53.8
Bon minori	040207	19	11	4	57.8	21.0

<sup>a</sup>Seeds compliments of Yasuo Ohta, D.Agr., D.Sc., professor, Tsukuba University in Japan.

Table 2. Agronomic characters of six soy varieties from Japan tested in provincial seed farm, Service of Agriculture, province of Kien Giang SR Vietnam - dry season 1981-1982

Entry number	Days to bloom	Days to maturity	Pubescence color	Flower color	Seed coat color	Hilum color	Seed size	No. of seed/pod
040691	25	70-75	white	purple	yellow	brown	medium	1-2
090125	25	70-75	white	purple	yellow	brown	small	1-2
090204	25	70-75	yellow	purple	yellow	brown	medium	2-3
040697	25	70-75	white	purple	yellow	brown	medium	1-2
040705	30	75-85	yellow	white	yellow	black	small	1-2
040207	30	75-85	yellow	white	yellow	black	small	1-2

100  
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247  
Notes on soybean nodulation with the indigenous rhizobium in Zambian soils [ ]

Introduction: The work reported here was carried out under the auspices of National Oilseeds Development Programme supported by the Research Branch, Department of Agriculture, Ministry of Agriculture and Water Development, Government of the Republic of Zambia, and the Food and Agriculture Organization of the United Nations.

In the 1977-78 season, the commercially grown soybean variety 'Hernon 147' was noticed to have nodulated profusely in a virgin land, without artificial seed inoculation. This was in the Mkushi area where soybean production then had just begun to expand. Observations made elsewhere in the country confirmed that only this variety, out of the five currently recommended, was capable of nodulating without seed inoculation. The effectiveness of nodulation was confirmed by excellent growth, dark coloured plants and commercial yields in excess of 2.0 tons per ha being reported by farmers. Experimental data recorded under best management practices, including seed inoculation prior to planting, confirms that the yield potential of this particular variety is not much higher than 2.0 tons per ha in that area.

In 1979-80, the same observation was made in the same area, for a breeding line under experiment and in a farm test program. The line was released in April, 1981, under the name 'Magoye' as the second commercial soybean variety with this ability to nodulate. Excellent growth and dark-colored plants were observed and commercial yields in excess of 3.0 tons per ha were reported by farmers, a yield very close to the variety's potential.

The significance of varieties capable of nodulating and producing high yields without seed inoculation is for the expansion of soybean production among the small scale farmers who will not have easy access to inoculum. The extension of information on soybean production to these farmers started in 1980.

Investigation: In order to assess the ability of various [varieties] for effective nodulation with the indigenous rhizobium in the Zambian soils, 400 varieties from the germplasm collection were planted at three sites, namely, Magoye, Lusaka and Mkushi, all in virgin land where land clearing was done just before planting. A compound fertilizer (10N: 20P205: 10K20: 10S) was applied at the rate of 300 kg per ha. Seeds of each variety were planted in single one-m rows, at 33 seeds per m, with a row spacing of 50 cm, without any seed inoculation.

No other management practice was carried out. The varieties were examined at the end of the flowering/beginning of pod formation for the following points:

- a. Plant color and growth;
- b. Number of nodules per 10 plants;
- c. Number of nodules red and pink per 10 plants;
- d. Nodules were samples for serological identification. Samples were sent to the NIFTAL Project at the University of Hawaii, but results have not yet been received.



By way of preliminary analysis, the varieties with more than 100 nodules per 10 plants and with more than 80% of nodules colored red or pink (as a sign of being active), were selected from each site. These numbered between 35-50 per site, among which there were varieties with acceptable agronomic characteristics.

This preliminary analysis indicated that, of those selected varieties for each site, all except a few (for which the genetical background is yet to be discussed with the original breeder) have a similar genetical background to that of the two varieties, Hernon 147 and Magoye.

The variety Hernon 147 was bred at Salisbury Research Station from a cross between Hernon(ex USA) and a "nonshattering" strain from South Africa, and was released in about 1940.

Variety Magoye was a cross between 'Gilbert' and 'K53', both of which were derived from a cross of 'Mamloxi' (CPI 1719 ex Nigeria) x 'Avoyellos' (CPI 15939 ex Tanzania). The cross was made at the University of Queensland by Dr. Don Byth and selection work was carried out in Zambia following provision of the breeding lines since 1975. The variety was released in April 1981.

#### Conclusion and future work

The results indicate that, under Zambian conditions, it is possible to select high yielding varieties with acceptable agronomic characters, capable of effective nodulation with the indigenous *Rhizobium*, from among the existing materials in the present germplasm collection.

Work will continue to select cultivars and test them with and without seed inoculation. It should be possible to use such cultivars in a breeding program. More studies are also required on the effectiveness of nitrogen fixation and utilization and it is hoped that this can also be undertaken.

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## VIII. INDEX OF AUTHORS

Agrawal, P.C. ....	35	Leudders, V.D. ....	135
Almeida, A.M.R. ....	17, 18	Mortimore, C.G. ....	30
Almeida, L.A. ....	18	Newhouse, K. ....	129
Almodiente, R.B. ...	97	Nyemba, R. ....	173
Anderson, T.R. ....	29	Palmer, R.G. ....	117, 140, 143
Ashawa, B.M. ....	58	Parot, C. ....	115
Bailey, Z.E. ....	109	Prakash, R. ....	33, 35
Beversdorf, W.D. ...	4	Pushpendra ....	39, 42, 43
Breithaupt, B.H. ...	131	Ram, H.H. ....	39, 42, 43
Buttery, B.R. ....	24, 26	Rana, N.D. ....	45, 47, 51, 53, 58, 62, 66, 68, 71, 75, 81
Buzzell, R.I. ....	23, 24, 26, 29, 30	Rao, M.S.S. ....	35
Chand, G. ....	75	Roth, J. ....	157
Chiang, Y.C. ....	140, 143	Sadanaga, K. ....	121, 123, 126, 129
Delannay, X. ....	121	Shanmugasundaram, S.	93, 95, 97, 99
Devine, T. ....	131	Sharma, S.K. ....	58, 75
Garg, I.K. ....	45, 47, 51, 53, 62 66, 68, 71, 75, 81	Shoemaker, R.C. ....	117
Haas, J.H. ....	30	Sichkar, V.I. ....	161, 163
Hiep, T.V. ....	169	Singh, J.M. ....	71
Hymowitz, T. ....	112	Singh, K. ....	39, 42, 43
Javaheri, F. ....	173	Smutkupt, S. ....	103
Kalia, R.K. ....	81	Soldati, A. ....	4
Keller, E.R. ....	4	Thieu, N.V. ....	169
Khanh, V.C. ....	169	Tin, C.H. ....	167, 168, 169
Kiang, Y.T. ....	140, 143	Toung, T.S. ....	97, 99
Kiihl, R.A.S. ....	17, 18	Trivedi, H.B.P. ....	33
Kovalchuk, M.V. ....	163	Tuan, T.T. ....	168
Kueneman, E.A. ....	87	Vaughn, D.A. ....	112
Lamseejan, S. ....	103	Verma, V.D. ....	39, 42, 43
Lark, K.G. ....	157	Walker, R.D. ....	23
Lau, D.V. ....	169	Wongpiyasatid, A. ..	103
Levitsky, A.P. ....	161, 163	Yen, C.D. ....	93, 95

245  
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